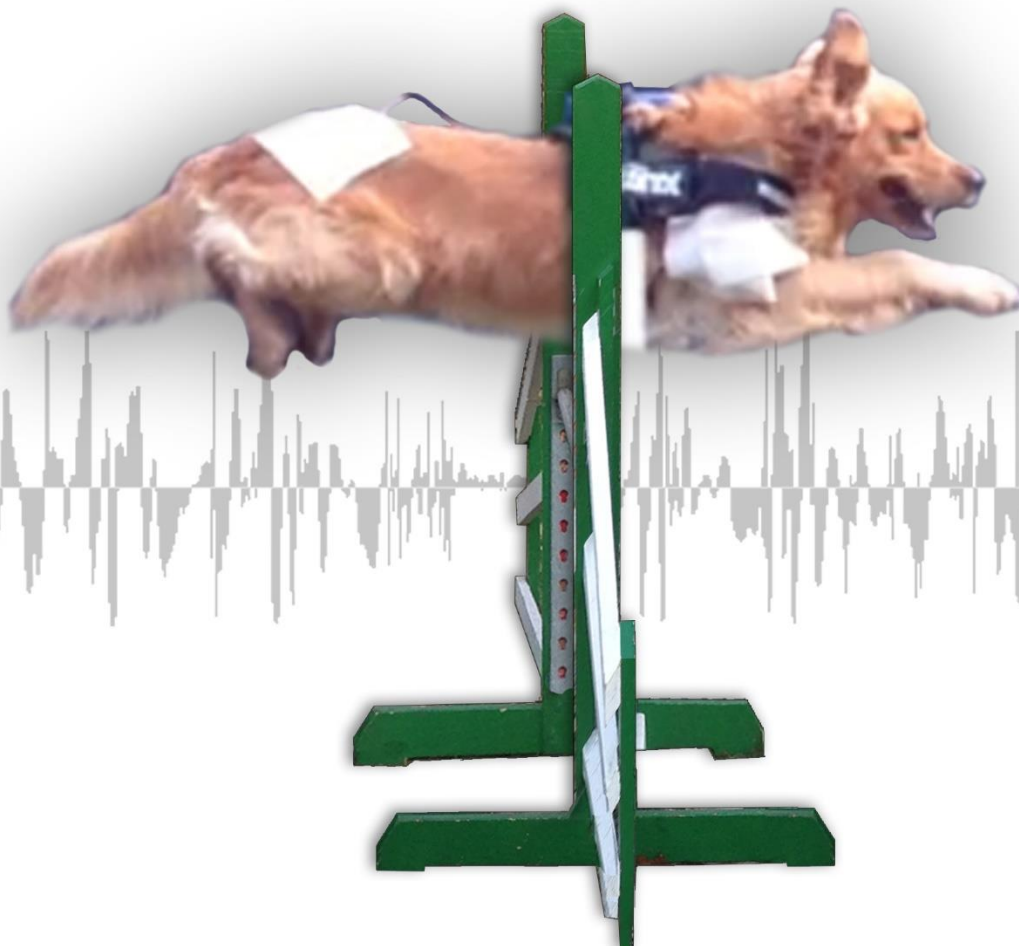




**Changes in muscle activation in large canines performing hurdles and A-frames in three different heights including warm-up and cool-down exercises**

- with the use of acoustic myography.

**Master Thesis in Animal Science by Lene Høeg Callesen, mpz419**



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## Preface

This master thesis has been performed to fulfill the requirement of a master degree in Animal Science and is nominated to 45 ECTS points. It was made at the University of Copenhagen in cooperation with the Department of Clinical Veterinary Medicine and Animal Science, Faculty of Health and Medical Sciences, University of Copenhagen, Denmark, in the period of October 2016 to August 2017.

This master thesis was supervised by Adrian Paul Harrison, who besides being associate professor at the University of Copenhagen, also has invented the acoustic myography CURO unit, which is used in the experimental party of this thesis.

This thesis contains a theoretical part leading to the experimental part and both has been sharply divided in four subjects: warm-up exercises, hurdles, A-frames and cool-down exercises to give a better overview of the project.

First of all, I would like to thank my supervisor Adrian Paul Harrison for sharing his knowledge with me and allowing me to use his invention. Thanks to Anne Désiré Vitger and Vibeke Sødring Elbrønd for standing by as co-supervisors. Thanks to the company MyoDynamik ApS for borrowing of the CURO and equipment, and from this opening a range of future possibilities within a field that interest me. I would also like to thank Kiara Salomons from MyoDynamik ApS for assisting during the many cold hours of measuring muscle activity in dogs and for sharing her knowledge. Thanks to Thomas Scheike and Torben Martinussen from Section of Biostatics, for much appreciated guidance with the statistical work. Finally, but not the least, I would like to give a special thanks to all the dog owners for participating in the experiment and allowing me to use their skilled dogs and for their hard work during the trials. Also thank to Royal Canin Nordics for sponsoring goodie bags to the participants.

Date: \_\_\_\_\_

Signature: \_\_\_\_\_

## Abstract

Canine agility has become one of the fastest growing dog competition sports with dog owners guiding the dog through a fixed course with hurdles, ramps and other obstacles in high speed. Dogs participating in agility is in risk of getting soft tissue injuries in their shoulders and backs, especially when running the A-frame and jumping hurdles. The aim of this thesis was to investigate how the muscles in dogs are affected by altered height of the hurdles and the A-frame to avoid agility-related injuries. Additionally, the effect of chosen contact method on the A-frame was investigated, comparing the running and the 2on-2off contact method. As the dogs were to be warmed up and cooled down in connection with the trials, the effect of warm-up and cool-down exercises on muscle function was also investigated.

Muscle activity was assessed with the use of acoustic myography which allows assessing the muscle function of dogs in real time while the dogs are performing agility. Fourteen large dogs of different breeds participated in 4 trials. First the dogs were warmed-up performing general and specific warm-up exercises for 3 times 5 minutes. Second the dogs jumped a sequence of 5 hurdles positioned 7 meters apart, first with a height of 45cm (low), next 55cm (medium) and finally 65cm (high). This was repeated 3 times. Thirdly the dogs ran the A-frame three times in three different heights 170cm (high), 150cm (medium) and 120cm (low) respectively while practicing their preferred contact method. Subsequently they ran the high A-frame three times using the other contact method. Fourthly the dogs performed a 15-minute cool-down exercise. During the four trials, the dog wore a harness equipped with a CURO measuring muscle activity in triceps brachii and gluteus superficialis on the right side of the dog via sensors positioned above each muscle. The dog owners were asked questions regarding the characteristics of the dogs, practice of agility as well as history of injury.

The results showed an increased coordination in triceps brachii after 5 minutes of warm-up exercises and a general improved use after 10 minutes of warm-up exercises. Meanwhile no effect was found in gluteus superficialis. The dogs generally had an improved use of triceps brachii and used gluteus superficialis more coordinated when jumping high hurdles compared to low. Dogs used to practice the 2on-2off contact method recruited less muscle fibres in gluteus superficialis when running up the low A-frame compared to the high. Meanwhile triceps brachii was used more coordinated when running down the high A-frame compared to the low when the contact method used was not considered. The dogs used triceps brachii more coordinated and overall better when practising the 2on-2off contact method compared to the running contact method when running both up and down the A-frame. Similar results were seen for gluteus superficialis when running down the A-frame. The cool-down exercise had no effect on muscle activity in the dogs. Dogs training weekly had an improved muscle activity during all exercises.

In conclusion, it cannot be recommended to lower the hurdles or the A-frame to avoid injuries and the practise of the 2on-2off contacts method is preferred. Dogs improve their muscle activity when performing warm-up exercises while cool-down exercises has no immediate effect. It is recommended to practise weekly as this improve muscle activity.

## Resumé

Hundeagility er en af de hurtigst voksende hundesports, hvor ejeren guider sin hund gennem en fastsat bane med spring, balance og andre forhindringer i høj fart. Hunde der deltager i agility er i risiko for at få skader i det bløde væv i skulder og ryg, specielt når de forcerer A-brættet og højdespringene. Formålet med dette speciale var at undersøge, hvordan hundenes muskler påvirkes af ændringer i højden på springene og A-brættet med det formål at undgå agilityrelaterede skader. Effekten af hvilken metode man har valgt til at få hunde til at tage felter på A-brættet blev også undersøgt ved at sammenligne løbefelter og 2på-2af felter. Da hundene alligevel skulle varmes op og køles ned i forbindelse med afvikling af forsøgene, blev effekten af opvarmning og nedkøling på musklerne også undersøgt.

Muskelaktiviteten blev undersøgt ved brug af acoustic myography, som gør det muligt at undersøge muskelfunktionen hos hunde i aktuel tid mens hunden løber agility. Fjorten store hunde af forskellige racer deltog i 4 forsøg. Først blev hundene varmet op ved at udføre både generel og specifik opvarmning i 3 gange 5 minutter. Dernæst sprang hundene en sekvens på 5 spring placeret med 7 meter imellem, først med en højde på 45cm (lav), dernæst 55cm (medium) og til sidst 65cm (høj). Dette blev gentaget 3 gange. I det tredje forsøg løb hundene over A-brættet 3 gange i 3 forskellige højder henholdsvis 170cm (høj), 150cm (medium), lav 120 cm (lav), mens de udførte deres vante feltræning. Efterfølgende løb de det høje A-bræt 3 gange, mens de udførte den anden metode til træning af felter. I det fjerde forsøg lavede hundene nedkølingsøvelser i 15 minutter. Under alle 4 forsøg bar hundene en sele med en CURO påmonteret. Denne målte muskelaktivitet i triceps brachii og gluteus superficialis på hundens højre side via sensorer placeret på huden over hver muskel. Hundenes ejere blev stillet spørgsmål omhandlende hundens karakteristika, agilitytræning og skadeshistorik.

Resultaterne viste en forbedret koordinering af triceps brachii efter 5 minutters opvarmning og en generel forbedring efter 10 minutters opvarmning. Til gengæld blev der ikke fundet nogen effekt på gluteus superficialis. Når hundene sprang de høje spring brugte de generelt triceps brachii bedre, og gluteus superficialis var bedre koordineret sammenlignet med når de sprang de lave spring. Hunde som var vant til at træne 2på-2af felter rekrutterede færre muskelfibre i gluteus superficialis, når de løb op ad det lave A-bræt sammenlignet med det høje. Til gengæld blev triceps brachii brugt mere koordineret, når de løb ned ad det høje A-bræt sammenlignet med det lave, hvilket gjaldt alle hundene uanset feltræning. Hundene brugte triceps brachii mere koordineret og generelt bedre når de udførte 2på-2af felter sammenlignet med løbefelter både på vej op og ned af A-brættet. Lignende resultater blev fundet for gluteus superficialis, når hundene løb ned af A-brættet. Nedkølingsøvelserne havde ingen målbar effekt på hundenes muskler. Hunde som træner ugentligt havde en bedre muskelaktivitet under alle forsøgende.

Det kan konkluderes, at det ikke kan anbefales at sænke højden på springene og A-brættet for at undgå skader, og at det bør foretrækkes at træne 2på-2af felter. Opvarmningsøvelser forbedrer hundenes brug af deres muskler, mens nedkølingsøvelser ikke har noget umiddelbar effekt. Det anbefales at træne ugentligt, da dette forbedrer brugen af musklerne.

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# 1.0 Introduction

## 1.1 Canine agility

Canine agility (agility) has become one of the fastest growing dog competition sports since its beginning as entertainment in the 1978 Crufts Dog Show and is today one of the largest canine activities in the world (Levy *et al.* 2013; Drinkwater 2017; United States Dog Agility Association 2017). In 2002 the American Kennel Club registered 239 trials with 106,661 entries, and after a total of 22 years offering agility trials, the numbers have continually increased until the newest statistics for 2016 with 3684 trials held with 1,223,660 entries (American Kennel Klub 2017). No such statistics can be found for agility competitions in Denmark, but the Danish Kennel Club (DKK) registered 37 trials in 2012 which has continually increased till 45 trials in 2016 (Dansk Kennel Klub 2017a). acknowledgement

Agility is a sport practiced by a handler with a dog. The handler shall guide the dog through a course with multiple obstacles within a predetermined time and the obstacles must be assessed in the correct order. The obstacles include high jumps (hurdles), A-frame, wall, tyre, see-saw, dog walk, flat tunnel, tube tunnel, weaving poles, long jump and table. The design of each obstacle can be seen in appendix A. There are different regulations all over the world, but to ensure a homologue execution there are some major international organizations who have made a set of rules or guidelines to follow. This will be explained further in section *1.1.2 Stakeholders*.

In Denmark, the dogs are divided into three heights small, medium and large respectively. Small is for dogs measuring less than 35 cm at the withers, medium is for dogs measuring between 35 cm and 43 cm at the withers, and large is for dogs measuring more than 43 cm at the withers (Federation Cynologique Internationale 2017a). There are three levels of complexity. The participants start at beginners' level at level 1, but as they run a certain number of trials without failure and within a restricted time they move up to level 2 for more challenging courses, and may advance to level 3 as the highest level. For each level, the height of hurdles is increased, while the height of the other obstacles is unaltered. The course may consist of up to 22 obstacles with no less than 7 hurdles. (Dansk Kennel Klub 2014; Danmarks civile Hundeførerforening 2015; Federation Cynologique Internationale 2017a). For 10 agility competition courses in DcH held in 2017 the hurdle was on average performed 13 times ranging from 11 to 14 hurdles on a course (Ehrenreich 2017). The repetitive nature of jumping hurdles when performing agility may contribute to a higher risk of injury (Levy *et al.* 2013; Pfau *et al.* 2010).

Agility is associated with a risk of injuries in dogs and as the sport has evolved over the years more focus has been made on security and avoiding injuries, both through studies and change of regulations.

### 1.1.1 Risk, site and cause of injury

Injuries can be classified according to which tissue is injured being skin, bone, muscle/tendon and ligament. However, this study will focus on the muscles in which strains, sprains and contusion may occur. In recent years studies have been made to investigate the risk and cause of injuries in dogs performing agility.

Levi *et al.* (2009) completed a preliminary retrospective survey in the period from November 2005 to February 2006 in which 1627 agility handlers answered a one-page questionnaire regarding agility related injuries in their dogs. As it is a preliminary survey it should be interpreted with caution.

Cullen *et al.* (2013a,b) performed a retrospective internet-based survey in the period from March to September 2009 in which 1669 handlers from all over the world completed surveys for 3801 dogs regarding agility related injuries in their dogs. From this survey two articles have been published, one examining the nature and perceived cause of injury in dogs and another using multivariable techniques to find risk factors.

Zachary *et al.* (2014) made a pilot study in which a questionnaire was administered to 217 handlers with 431 dogs competing in 5 trials in Ohio over a 4-month period and ending with a follow-up survey at a sixth trial. The questionnaire regarded agility-related injuries among both handler and dogs, but only the results for the dogs is of interest in this thesis. As it is a pilot study with a small sampling size, it should be interpreted with caution.

For the purpose of getting an easier overview of the results from the different surveys a table is made listing the risk, type, site, cause and severity of agility-related injuries. This can be seen in table 1 in which the two articles by Cullen *et al.* 2013a and -b are handled as one source.

Dogs get injured both at training and competition, but taken the hours spent on performing agility into account a tendency for a higher risk of injury at competition is shown. Border collies are at higher risk of injury and alternative therapeutic treatments as well as the experience of the dog and handler may have an influence of risk of injury. It is often soft tissue injuries in shoulder and back caused by direct contact with A-frame, dog walk or hurdles. It results almost equally in both minor and major injuries. (Levy *et al.* 2009; Cullen *et al.* 2013a,b; Zachary *et al.* 2014)



A risk of injury is inevitable when performing agility, but what can be done to minimize the risk. Both agility related organizations and the handlers as stakeholders are making an effort to lessen the risk of injury.

Table 1a: Results found in four articles: Levy *et al.* (2009), Cullen *et al.* (2013a,b), Zachary *et al.* (2014) respectively describing risk, type, site, cause and severity of agility-related injuries.

Article	Levy <i>et al.</i> 2009	Cullen <i>et al.</i> 2013a,b*	Zachary <i>et al.</i> 2014
Risk	33% of the dogs has been injured, of these 58% occurred during competition while the 42% occurred during training. Border collies were significantly more often injured.	32% of the dogs had been injured. The risk of injuries is the same for competition (46%) and training (45%). Dogs are at higher risk when: they have previously been injured, dog have $\leq 4$ years of experience and the use of alternative therapeutic treatments. Contrary dogs are at lower risk when: dog have $>4$ years of experience and handler has $\geq 5$ years of experience.	Of the injured dogs (n=35) 45% occurred during competition (1.74 injuries per 10000 runs) while 47% occurred during training (1.72 injuries per 10000 hours).  Dogs visiting a chiropractor had 2.63 times odds of injury and dogs getting massage had 2.40 odds of injury.
Type	Soft tissue injuries were the most common type of injury. Sprains, strains and contusions.	Soft tissue injuries were the most common type of injury. Sprains, strains and contusions.	Strains, cut/scrapes and sprains
Site	The shoulders (20%) and backs (18%) were most commonly injured. The stifle, hip, carpus, phalanges were injured less frequently.	Shoulder (23%), back (19%), phalanges (13%), neck (12%) A significant higher number of injuries was found for: - hurdles in: shoulder, stifle joint, carpal joint and antebrachium. - the A-frame in: shoulder and phalangeal. - the dog walk in: ribcage and head.	NA
Cause	Direct contact with an obstacle (35%), contact with ground (18%), slips (18%), overuse (17%) The three obstacles causing the highest number of injuries where A-frame (29 %), dog walk (19%) and hurdles (17%).	The three obstacles causing the highest number of injuries where hurdles (16%), A-frame (15%), dog walk (11%) Environmental factors (eg. wet grass) (13%)	Direct contact with an obstacle (32%), slips/fall (incl. obstacles) (29%), The four obstacles causing the highest number of injuries where A-frame (11%), flat tunnel (11%), tyre (not a break away tyre)(8%) and dog walk (8%).
Severity	For obstacle-related injuries 46% caused minor injuries and 42% caused major or chronic injuries. 67% of the retired dogs retired due to agility-related injuries.	For the injuries occurring at practice 55% were minor and 45% were severe injuries. For the injuries occurring at competition 52% were minor and 48% were severe injuries.	26% were minor injuries, 26% were major injuries and 18% of the injuries were chronic while 5% required surgical repair.

\*Cullen *et al.* 2013a and -b are listed as one source as the two articles come from the same survey.

### 1.1.2 Stakeholders

In Denmark, most associations offering agility follow the rules made by the Fédération Cynologique Internationale (FCI). The FCI is a world canine organization with 91 member countries and a contract partner for each country. Besides making sure the judgement of pedigrees is homologue, FCI also set a standard for international agility competitions to ensure a homologue execution between member countries. (Federation Cynologique Internationale 2017b). This is done through agility regulation, equipment standards and guidelines for agility judges made by the FCI (Federation Cynologique Internationale 2011a; Federation Cynologique Internationale 2017a). These regulations are made and revisited by the FCI agility committee with experts from member countries, where Allan Hansen who is the chairman for agility in the Danish Kennel Club (DKK) currently represent Denmark (Hansen 2017). The Danish Kennel Club is the contract partner in Denmark and full member of the FCI. The DKK primarily focus on the work with pedigrees, but do also offer activities for dogs and their owners such as agility (Dansk Kennel Klub 2017b). Besides DKK other bigger associations such as Danmarks civile Hundeførerforening (Denmark's civilian dog handler association (DcH)) and Danske Gymnastik- og Idrætsforeninger (Danish Gymnastics and Sports Associations (DGI)) offer canine agility training and competitions together with other smaller associations (Danske Gymnastik- og Idrætsforeninger 2017b; Danmarks civile Hundeførerforening 2017a; Kvistgård Agility Forening 2017; Dansk Agility Forening København 2017). Most of the agility associations in Denmark follow the FCI regulations for a homologue practicing of the canine sport in the country, although some with modifications as suggested by the FCI (Dansk Kennel Klub 2014, Danmarks civile Hundeførerforening 2015, Danske Gymnastik- og Idrætsforeninger 2017a, Federation Cynologique Internationale 2017a).

Both the FCI, DKK, DcH and DGI make the regulations in the interest of the participants and the security of the dogs. The participants may affect the work of the organisations through public discussions which has been more pronounced in recent years as Facebook has offered a large advantage on this part (Sommer & Berthelsen 2017). This has given the participants more power, but also made room for all kind of rumours regarding speculations of changes in rules and fast distribution of unscientific knowledge of what causes injuries. The judges have a great deal of power themselves as they may choose to avoid using obstacles they deem to be unsecure and make a safer course for the dogs as they approach and leave the obstacles. An example could be the case with the closed tunnel. In recent years, the closed tunnel has been highly discussed, due to the risk of the dogs slipping and getting entangled in the closed tunnel as well as bruising their noses against the fabric. Pressure from the participants has forced the agility committees in several countries to reassess the equipment. From August 30, 2016, the American Kennel Club and The Kennel Club in UK has suspended the use of the closed/collapsed tunnel due to safety of the

dogs (The Kennel Klub 2016; Herwig 2017). They are not members of the FCI and have their own council regulating the rules. Shortly after the suspension of the closed tunnel in the American Kennel Club and The Kennel Club in UK there have been no or very little use of the closed tunnel in Denmark chosen by the judges. Although the closed tunnel is currently not going to be omitted, the design and use will be revisited (Hansen 2017). Generally, the regulation and the obstacles have been revisited over the years to avoid injuries.

### 1.1.3 Change of regulation and practise to avoid injuries

To avoid injuries and in the interest of evolving the sport, adjustments have been made to the agility regulations.

In 2014 the DcH changed the regulation according to the height of the hurdle. Previously the heights for the large dogs was 65 cm, but with the newest regulation an interval from 60-65 cm was made which is similar to the level 2 height (Danmarks civile Hundeførerforening 2014). This opened the opportunity for lowering the heights of the hurdles in level 3 if chosen by the judge. This also apply to the current regulation from the DKK. Still the height of the hurdles is most often higher in level 3 compared to level 2.

For the FCI the most recent regulation was made in January 2012 where the plank of the dog walk was changed to being no longer than 3.80 meters and the height no longer than 1.30 meters compared to the previous measures of 4.20 meters and 1,35 meters respectively (Danske Gymnastik- og Idrætsforeninger 2017a, Federation Cynologique Internationale 2017a). This resulted in a higher slope on the plank going up and down the dog walk for many dog walks in Denmark. Likewise, the length of the see-saw has been shortened from being no longer than 4.2 meters to 3.8 meter (Danske Gymnastik- og Idrætsforeninger 2017a, Federation Cynologique Internationale 2017a). This is probably done to minimize the force of impact, but the reason for this change is currently unknown, as there has been no publication from the FCI agility committee (Hansen 2017). The height of the A-frame was lowered from 1.9 meter to 1.7 meter approximately 10 years ago (Hansen 2017), but no documentation regarding this was found. A lot of the reasoning behind the change of regulations is not published (Hansen 2017). In march 2007 the United States Dog Agility Association (USDAA) changed the height of the A-frame for large dogs to be no higher than 180 cm (71 inches) compared to the earlier 190 cm (75 inches) (United States Dog Agility Association 2007). This was among others done to lessen the impact of dogs running the A-frame with the 2on-2off (2o2o) contact method (Schwarz 2007).

The 2on-2off is a method to ensure the dogs touches the contact area which is the last 106 cm from the bottom of each ramp on the A-frame (picture 1) and 90 cm on the dog walk and see saw. This area is painted in another colour than the rest of the obstacle and shall be touched with at

least one paw (Federation Cynologique Internationale 2017a). The exception is the run up the A-frame at the DcH (Danmarks civile Hundeførerforening 2015). This rule is made for security reasons ensuring the dog do not jump from high ground. There are two main practices in Denmark when training the insurance of taking the contact area, the 2o2o-contact method and the running contact method. When practising the 2o2o-contact method the dog runs the A-frame and before leaving it stands still with its hind paws on the contact area and the front paws on the ground until a release command is given (picture 1). When using the running contact method, the dog touches the contact area in the run and do not slow down (picture 2). Both methods may be trained with strict methods or loosely learned, and the results and efficiency differ greatly at competitions. The 2o2o-contact method is highly discussed due to the impact force on the legs when making the stand, which is one of the reasons, together with the increased speed, some handlers prefer the running contacts.



Picture 1: Dog practicing 2on-2off contacts on an A-frame.



Picture 2: Dog practicing running contacts on an A-frame.

The Federation Cynologique Internationale (2017c) has published a new regulation valid from January 2018 in which the maximum height of the hurdles will be lowered 5cm in all sizes resulting in a maximum height for the large dogs of 60cm. How this may affect the dogs and the risk of injury is unknown. For the FCI, DKK DGI and DcH to make scientific evidence supported changes in regulations they need scientific knowledge of the physics and anatomy of the dogs and how the changes in regulation may affect them to achieve the actual goal of avoiding injuries.

#### 1.1.4 A sum-up

Dogs performing agility are at increased risk of getting soft tissue injuries in shoulder and back especially when jumping high hurdles or running the A-frame. Studies have been made to find the cause of injury and to investigate how the dogs are affected by different setups. Still requests are made for further studies to identify how the internal limb structures may be related to injury (Pfau *et al.* 2010), and cited from Cullen *et al.* (2013b) “An important future goal is to identify improvements on equipment, techniques, or both that may reduce the risk of injury among these

*canine athletes*” This must be done not only in the interest of the organizations making the rules, but also in the interest of the handlers performing agility with their dogs.

The hurdles and A-frame will be in focus in this thesis together with the muscles triceps brachii and gluteus superficialis. The reason to choose these muscles is that the activity of triceps brachii are increased when both ascending the A-frame and jumping hurdles (Cullen *et al.* 2017), and gluteus superficialis make propulsion work together with gluteus medius (Robert *et al.* 2000). From this knowledge, it is expected that these muscles are affected when jumping hurdles and running the A-frame. Their activity can be measured with the use of acoustic myography (AMG) showing in ESTi<sup>TM</sup>-scores and this will be further explained in section 1.8.1 *Acoustic myography*.

## 1.2 Problem formulation and hypothesis

The purpose of this thesis is to investigate how the muscles in dogs are affected by altered height of the hurdles and the A-frame to avoid agility-related injuries.

This is investigated with the following hypotheses:

1. Triceps brachii and gluteus superficialis shows a lower level of muscle engagement when jumping lower compared to higher hurdles, as assessed by a higher ESTi<sup>TM</sup>-score.
2. Triceps brachii and gluteus superficialis shows a lower level of muscle engagement when running an A-frame with a lower height and slope compared to a higher and steeper A-frame, as assessed by a higher ESTi<sup>TM</sup>-score.
3. Triceps brachii and gluteus superficialis shows a lower level of muscle engagement when using a running contact method compared to the 2on-2off contact method when running the A-frame, as assessed by a higher ESTi<sup>TM</sup>-score.

As it is intended to make the dogs perform warm-up and cool-down exercises before and after the trials respectively, the effect of this is measured as well, with the following hypotheses.

4. Triceps brachii and gluteus superficialis show a higher ESTi<sup>TM</sup>-score as a result of the performance of warm-up exercises.
5. Triceps brachii and gluteus superficialis show a higher ESTi<sup>TM</sup>-score as a result of the performance of cool-down exercises.

The reasoning behind these hypotheses’ will be described in the following sections.

### 1.2.1 Delimitations

Some delimitations have been applied to the thesis to minimize the size of the thesis, but also due to more practical reasons. No more than two agility obstacles, each in three heights, were chosen

due to the time available and to avoid muscle fatigue. The number of muscles are chosen to limit the size of the thesis, but also as a consequence of the chosen assessment technique. The AMG-equipment also limited the size of the dogs being able to participate in the experiment as explained in section 2.1 *Recruitment of participants*, resulting in dogs below 43 cm at the withers were excluded. Only dogs from Denmark and especially the capital area would participate in the experiment due to the location of the trials. The effect of warm-up exercises on the performance of the following exercises will not be investigated. Likewise, the effect of the cool-down exercises will not be assessed hours and days after the trial. In general, there will be no comparison between the different type of exercises or between the two muscles. The kinetics, force and change of length of the muscles and other aspects of muscle work will not be assessed. Elastic energy will only shortly be mentioned and discussed in this study, as discussing the results with this angle would be a study by itself. Likewise, the speed of the dogs, if they stretch or flex their hip when jumping, number of steps between hurdles, type of gallop, strong and weak side, jumping technique etc. will not be assessed in this study.

## 1.3 Nerves

This thesis focuses on the muscles of the dog which consist of up to 40-45 % of the total bodyweight of the dog (Huard *et al.* 2002). Muscle fibres contract when a signal is received from a nerve fibre. A nerve fibre consists of dendrons formed by dendrites, which is short branched extensions, that carry nerve impulses toward the cell body where the nucleus is located (Sjaastad *et al.* 2010; Fisher & Lilje 2014). From here the impulses are carried away from the cell body by a single axon. The axon terminates in larger branches with nerve endings. When the nerve is connected with a muscle fibre it is called a neuromuscular junction and the synaptic cleft lies between the membranes (Sjaastad *et al.* 2010; Fisher & Lilje 2014). From here the nerve impulse stimulates contraction of the muscle fibres.

### 1.3.1 Activation of muscle fibres -Part I

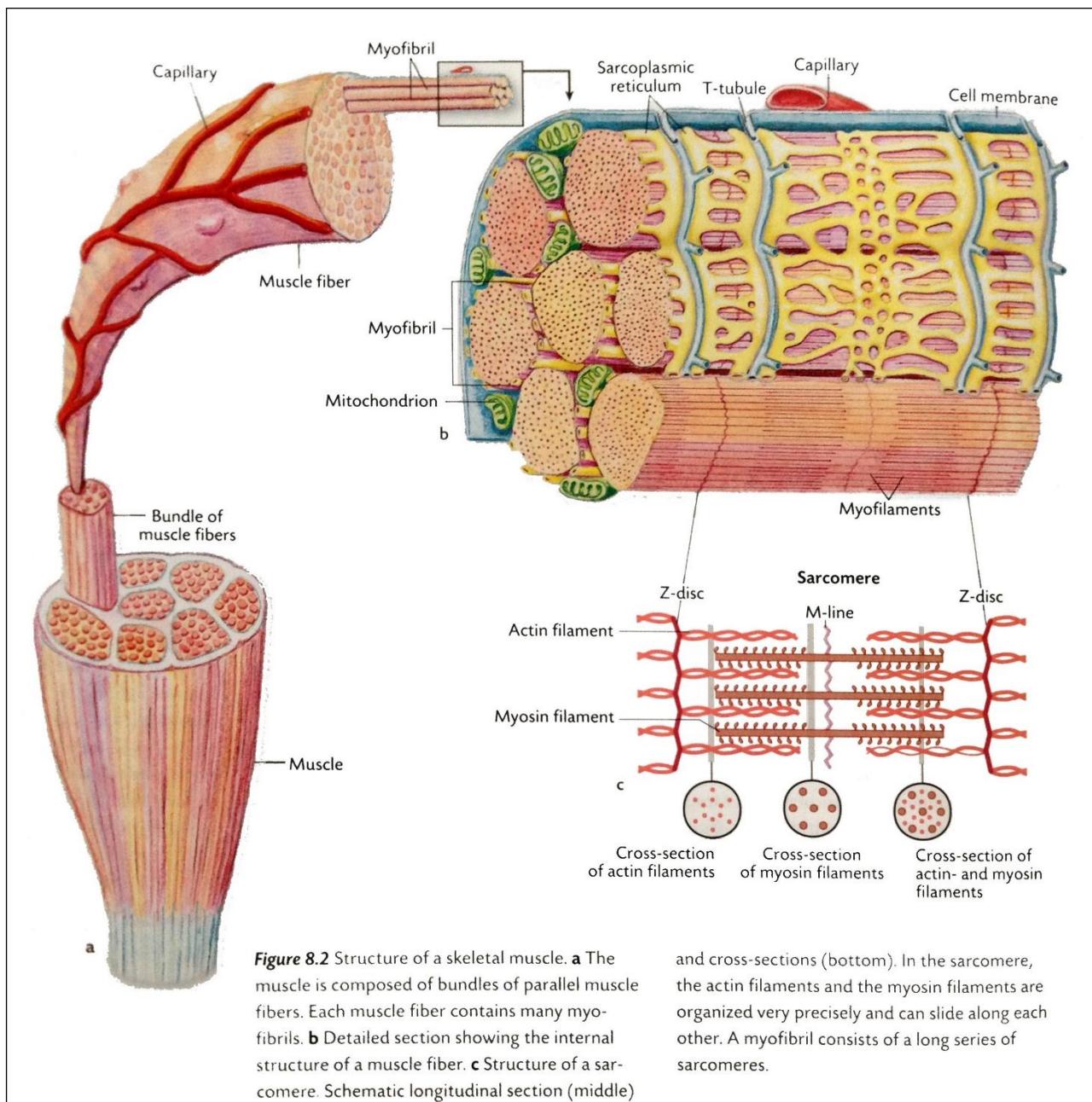
Neurons carrying impulses away from the central nervous system (CNS) are called motor neurons. (Aspinall & Cappello 2009; Sjaastad *et al.* 2010) Motor neurons send signal from the motor cortex of the brain to the lower motor neurons in the spinal cord. From here two types of motor neurons proceed alpha and gamma respectively. Together with the muscle spindle, which register the level of contraction, the alpha neurons innervate the muscles while gamma neurons innervate the muscle sensors and inform the spindle control group of changes in reference values. The muscle spindle sends information about changes in length of the contraction, and the alpha and gamma neurons respond accordingly (Sjaastad *et al.* 2010; Fisher & Lilje 2014). Thus, the muscle contraction may increase or decrease as needed. One motor neuron may activate up to thousands



of muscle fibres, all of the same type. Muscle fibres activated by the same motor neuron are called a motor unit and are all activated by the same time. The number of muscle fibres activated by the motor neuron is dependent on the precision of the work needed. A motor neuron activating few muscle fibres result in fine movements, while activation of thousands of muscle fibres results in gross movements (Aspinall & Cappello 2009; Sjaastad *et al.* 2010; Fisher & Lilje 2014). According to the work of the muscles additional or fewer motor units may be recruited or the signals may be send with a higher or lower frequency through the motor neuron resulting in a changed firing rate and amount of active muscle fibres recruited. (Harrison *et al.* 2017; MyoDynamik ApS 2017). This knowledge can be applied when measuring muscle function and will be further explained in section 1.8.3 *Interpretation of the ESTi<sup>TM</sup>-scores.*

## 1.4 Muscles

Muscles are composed of bundles of muscle fibres (Sjaastad *et al.* 2010). Each muscle fibre has several nuclei (myonuclear) and more myonuclear may be added by fusion of satellite cells when hypertrophy occurs (Sjaastad *et al.* 2010; Bazgir *et al.* 2017). Satellite cells is mononuclear muscle stem cells which have self-renewal and multi-differentiation capabilities (Bazgir *et al.* 2017). They lie between the basal lamina and plasma membrane and the number decreases with age (Bruusgaard *et al.* 2010; Bazgir *et al.* 2017). Each muscle fibre consists of multiple myofibrils which is composed of actin and myosin filaments (Sjaastad *et al.* 2010). These myofibrils lie in bundles and form units of contraction called sarcomeres. In one end, the actin filaments are attached to a protein lattice called the Z-disc, while the myosin's are attached to a protein lattice which is called the m-line. For visualisation and further details see picture 3 from Sjaastad *et al.* (2010). The largest of all known proteins, titin, is attached to the M-line and Z-disc and ensures that the myosin filaments are kept centered (Lindstedt *et al.* 2001; Sjaastad *et al.* 2010).

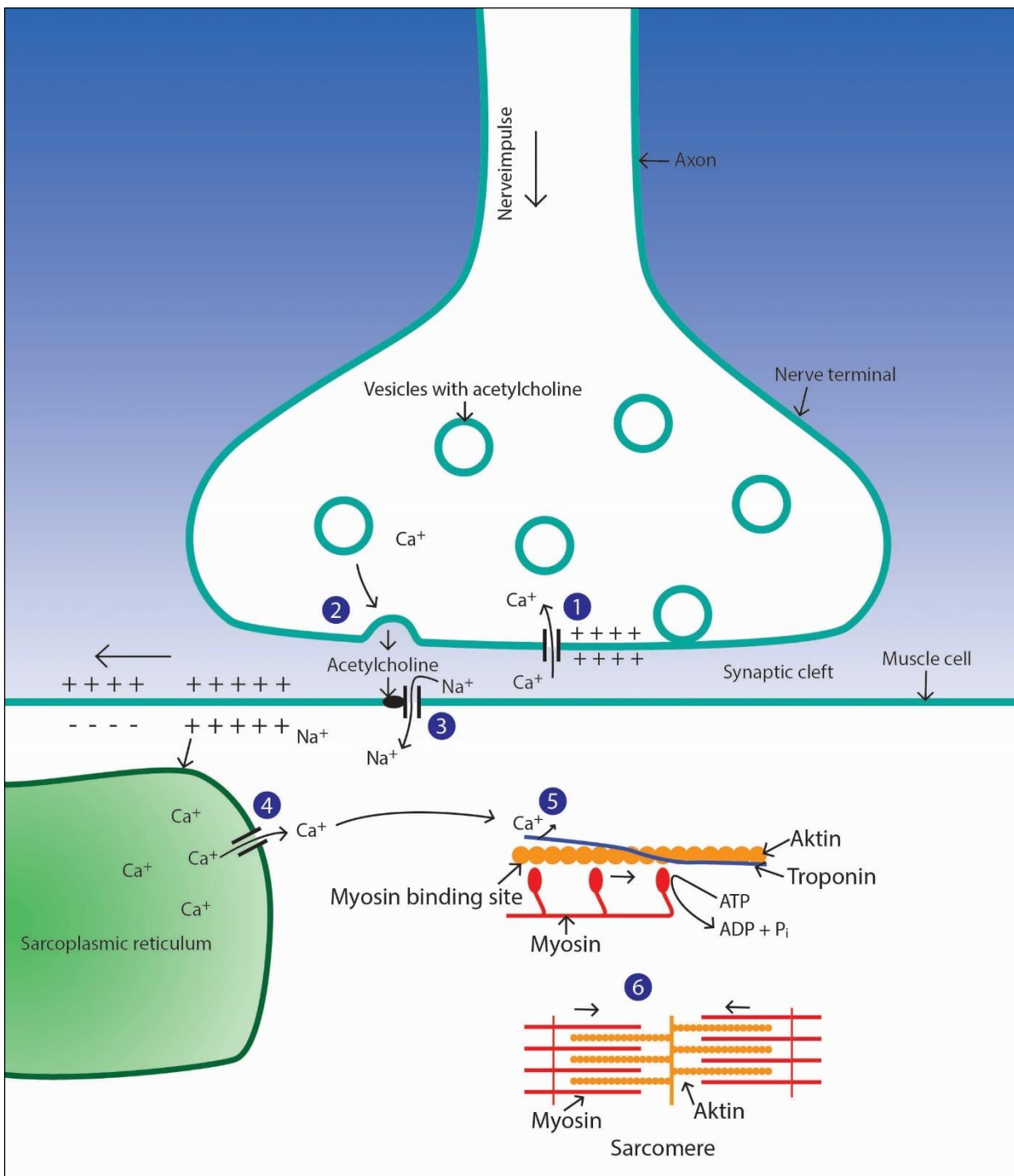


Picture 3: Illustration of muscle fibre composition. See original figure text in the picture taken from Sjaastad et al. (2010), page 282.

#### 1.4.1 Activation of muscle fibres -Part II

Muscles contract when the nerve terminal of the motor neuron that innervate the muscle is depolarized. For a better understanding of the underlying processes figure 4 is made with inspiration from Aspinall & Cappello (2009) and Sjaastad *et al.* (2010). Muscle contraction need energy supplied as adenosine triphosphate (ATP). Adenosine triphosphate can be obtained either by aerobic metabolism (respiration) or anaerobic metabolism (glycolysis) (Hopkins 2006; Aspinall & Cappello 2009; Sjaastad *et al.* 2010). This is dependent on the type of work and hence the muscle fibre type activated.





Picture 4: Illustration of muscle contraction. When the motor neuron is activated the nerve terminal is depolarized and calcium-ions enters the terminal end of the axon (1) and acetylcholine is released into the synaptic cleft (2). Here it is bound to receptors on the membrane on the muscle fibre resulting in a potassium influx (3) which depolarise the membrane of the muscle fibre and the cytoplasm. This induces calcium-ion release from the sarcoplasmic reticulum (4), and the increased calcium-ion concentration allow the myosin heads to bind to actin by removing troponin from the myosin binding site (5). Consequently, the sarcomeres shorten as the heads of the actin and myosin filaments pulls the filaments along each other using cross bridges, resulting in muscle contraction (6). This process need energy supplied as adenosine triphosphate (ATP). When calcium is retrieved to the sarcoplasmic reticulum the troponin covers the myosin binding site. The myosin heads release the cross bridges and the muscle relaxes. The drawing is made with inspiration from Aspinall & Cappello (2009) and Sjaastad *et al.* (2010).

### 1.4.2 Muscle fibres

Muscle fibres are divided according to their contractile, metabolic and morphological features. Four pure types of muscle fibres exist type I, IIA, IIB, IIX respectively and three hybrids type I-IX, IIA-IIX, IIB-IIX (Acevedo & Rivera 2006; Toniolo *et al.* 2007).

Type I muscle fibres have a diameter of 1. 1-2.4  $\mu\text{m}^2$  (Newsholme *et al.* 1988) and are slow oxidative fibres meaning they contract slowly by the use of oxygen. They are also called red muscle fibres due to the high myoglobin content which store oxygen within the muscle cell, but they only have small glycogen deposits. The design of the type I muscle fibres means they are often the first to be recruited for more continual work, and they are fatigue resistant (Hopkins 2006; Fisher & Lilje 2014). They are used to stabilize and maintain posture (Sjaastad *et al.* 2010; Fisher & Lilje 2014), and stabilizing muscles or part of the muscles close to the bones are often redder due to the higher content of type I fibres (Fisher & Lilje 2014).

Type II muscle fibres have a diameter of 1.9-3.1  $\mu\text{m}^2$  (Newsholme *et al.* 1988) and are further divided into type IIA and type IIX (Toniolo *et al.* 2007; Fisher & Lilje 2014). Equal for both of them is they are fast oxidative and glycolytic fibres, meaning they contract fast by the use of either oxygen or glucose. They are also called white muscle fibres and lack the red colour due to the less abundant content of myoglobin, but they do have a higher glycogen content. The design of the type II muscle fibres means they are used for short intensive work and are easily fatigued especially the glycolytic fibres (Hopkins 2006; Sjaastad *et al.* 2010; Fisher & Lilje 2014). They are used in jumping exercises and generally where high power of short duration is needed, and is abundant in muscles doing propulsive work (Sjaastad *et al.* 2010).

Humans have both type IIA and type IIB muscle fibres. Type IIA is both oxidative and glycolytic while type IIB is only glycolytic. Although type IIB muscle fibres have been seen in laryngeal and extraocular muscle in two large dogs (Toniolo *et al.* 2007), type IIB muscle fibres have not otherwise been found in dogs. Instead they have type IIX muscle fibres, which is oxidative and glycolytic, and according to contractile, metabolic and morphological features are an intermediate between type I and type IIA (Acevedo & Rivera 2006; Fisher & Lilje 2014). It is thought to be the explanation of the extraordinary levels of stamina in dogs (Acevedo & Rivera 2006). Type IIX muscle fibres can be found in the leg muscles, but are not found in some back muscles (Acevedo & Rivera 2006) nor diaphragm (Toniolo *et al.* 2007). Type IIX muscle fibres are especially frequent in semimembranosus and tibialis cranialis (Toniolo *et al.* 2007). Hybrids between IIA and IIX or IIB and IIX do also exist and IIA-IIX hybrids are abundant in longissimus dorsi (Toniolo *et al.* 2007). Pure IIX or the hybrids have previously been misclassified and associated as one muscle fibre type (Acevedo & Rivera 2006; Toniolo *et al.* 2007).

The distribution of type I fibres has been investigated in three mongrel dogs (Armstrong *et al.* 1982) and it was found that deep muscles in forearm and leg that serve to resist gravity had the highest percentage on type 1 fibres compared to the superficial muscles in the same group. In general, the deeper areas of the muscles had more type I fibres compared to the superficial areas (Armstrong *et al.* 1982).

No breedspecific differences in muscle fibre type composition have yet been found (Newsholme *et al.* 1988; Agüera *et al.* 1990 cited in Fisher & Lilje 2014; Toniolo *et al.* 2007), except for greyhounds which have a higher proportion of type II fibres (Gunn 1978). The composition of fibre type is not different between male and female in dogs (Gunn 1978), but it is discussed whether or not training may influence the fibre type composition.

#### 1.4.2 Muscles and training level

According to Gunn (1978) and Fisher & Lilje (2014) the muscle fibre type does not change according to training level. Dogs muscles are relatively immature at birth and develops until a mature morphology are reached at an age of two months (Strbench 2006). Puustjärvi (1994) found that 10 beagles trained on a treadmill from the age of 15 to 70 weeks had a higher percentage of type I fibres in triceps and a lower percentage in the lumbar multifidus muscles compared to a control group which had received no exercise. It should be noted that the percentage of type II fibres decrease with advancing age in male dogs (Braga *et al.* 2016). Alway *et al.* (2002) compared how exercise affected the muscles of young and aged rats. They found that exercises did not change the composition of muscle fibres in aged rats, but in young rats the content of type I fibres was significantly lowered. Muscles in elderly animals is less affected by overload, and like dogs aged rats has a lower content of type II fibres (Alway *et al.* 2002). In humans, the percentage of type II fibres is found to decrease during heavy-resistant-training, but bounces back to normal level when detrained (Staron *et al.* 1991). Further research is necessary to investigate the effect of training both in relation to the age of the dog and the level of exercise, however exercise has with no disputation an effect on the size of the muscles (Fisher & Lilje 2014). Heavy resistance exercise will result in an increase of myofibrils, myonuclear and satellite cells and a decrease in volume density of mitochondria (Lüthi *et al.* 1986; Kadi and Thornell 2000). Meanwhile, endurance training increases the capillary and mitochondria volume density (Hoppeler *et al.* 1985 cited in Hoppeler & Flück 2002). The muscle gets atrophied when not in use meaning they become smaller as the myofibrils is degenerated (Sjaastad *et al.* 2010; Fisher & Lilje 2014). Training alters the expression of gene isoforms mainly associated with the oxidative ATP production and express novel transcripts in humans (Lindholm *et al.* 2016). Studies of an overloaded muscle in mice has found that an elevated number of nuclei persisted after 2 years of detraining and that only 1% of the myonuclear is lost as a result of apoptosis (Bruusgaard *et al.* 2010). This may result in a more

efficient response to retraining (Bruusgaard *et al.* 2010), which has been found in humans (Staron *et al.* 1991), although proof of muscle memory is not found in humans on gene level (Lindholm *et al.* 2016).

### 1.4.3 Elastic energy

When force exerted on the muscle exceeds the force developed by the muscle, the muscle must absorb the mechanical energy also called negative work (Lindstedt *et al.* 2001). The absorbed energy may either be dissipated as heat or momentarily stored as elastic potential energy. Elastic potential energy is the energy that are stored when a material is deformed, like the tendons or muscles when jumping which helps maximizing jumping power (Gregersen & Carrier 2004). Elastic energy can be stored in structures outside the muscle fibres such as collagen and tendons, but may also be stored in the titin inside the fibres (Lindstedt *et al.* 2001). The stored energy is recoverable and can be added to the subsequent stride, making the pace more energy efficient (Gregersen *et al.* 1998; Lindstedt *et al.* 2001). Thus, the pre-stride before jumping is important both to prepare for the correct distance, angle and speed, but also to store elastic energy to add to the power needed to jump the hurdle or ascending the A-frame. Dogs may not store energy as efficient as horses as their paws functions as shock absorbers (Williams *et al.* 2008a). Still, dogs have been found to store high amounts of strain energy in their Achilles tendons when jumping long jumps (Alexander and Bennet-Clark 1977), and the front limbs has an even greater potential for storing elastic energy when trotting and galloping (Gregersen *et al.* 1998). Generally, more storage of elastic energy is produced in the distal extensor muscles when trotting and galloping compared to the more proximal extensor muscles (Gregersen *et al.* 1998).

## 1.5 Types of muscle contraction

Muscles undergo two types of contraction the isometric and the isotonic contraction respectively. In the isometric contraction, the muscle contract without changing length e.g. to hold posture while additional weight is put on. Opposite the isotonic contraction occurs when the muscles are either shortening (concentric) or lengthening (eccentric) (Enoka 1996; Aspinall & Cappello 2009; Gillette & Dale 2014). Abbott *et al.* (1952) found that a greater muscular force was needed for eccentric exercises compared to concentric exercise. In general, the energy cost for eccentric contractions are low and the contraction more efficient, while the magnitude of the force is high (Lindstedt *et al.* 2001; Lauer *et al.* 2009). It is discussed how the nervous system activate the muscles during eccentric contraction. It is postulated that a unique activation strategy is needed as the actin and myosin do not slide against each other with the consumption of ATP, but the bonds are disrupted mechanically during eccentric contractions (Enoka 1996).

### 1.5.1 Eccentric muscle contractions related injuries

Due to the high force, eccentric contractions often cause muscle damage, especially when a shift in the normal pattern occurs, but with proper training injuries can be avoided (Lindstedt *et al.* 2001). In humans, injuries in hamstring muscles most often occur in the swing phase of the gait as a consequence of eccentric contraction, and repeated movements may result in accumulation of macro damages that predispose to muscle injury (Schache *et al.* 2009; Schache *et al.* 2010; Chumanov *et al.* 2007). A higher number of type II fibres are recruited under eccentric exercises (Nardone 1989) and injuries are more common in fast fibres with low oxidative capacity (Friden & Lieber 1992). It is suggested that type I fibres are less vulnerable than type II fibres due to a greater actin and myosin cross bridge called Z-discs, making them more resistant to injuries related to high tension and exercises with a repetitive nature (Friden & Lieber 1992). The eccentric work weakens when the muscle has previously been damaged and the muscle are more prone to damage from eccentric exercise (Jonhagen *et al.* 1998; Brockett 2004).

## 1.6 Anatomy and biomechanics

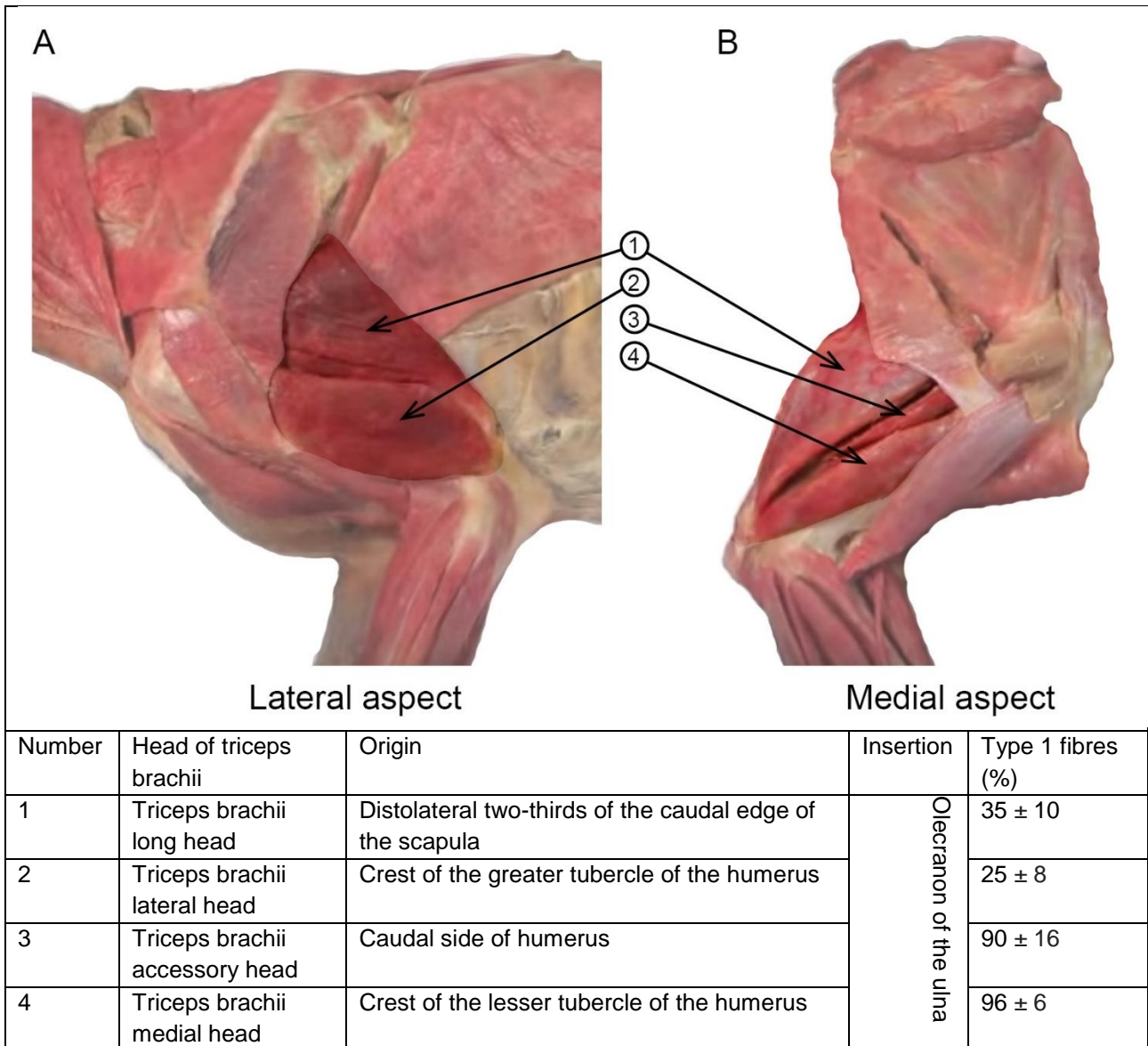
The muscles in focus are triceps brachii and gluteus superficialis. These muscles are chosen due to both anatomical and physiological reasons as well as practical reasons according to the AMG-equipment used as described in section 2.2 *Pilot study*.

### 1.6.1 Triceps brachii

Triceps brachii has four heads the long-, lateral-, medial- and accessory head respectively and each with separate origins. They have a common insertion point on the olecranon of the ulna. The nerve radialis innervate triceps brachii (Miller 1993) and when contracting the muscle extend the elbow joint (Miller 1993; Williams *et al.* 2008b; Fisher & Lilje 2014) and act as shoulder flexor (Williams *et al.* 2008b; Cullen *et al.* 2017). Triceps brachii and the position of the four heads and their content of type I muscle fibres are illustrated in picture 5 and listed in the accompanying table. The long head of triceps brachii works as both an elbow extensor and a shoulder flexor due to its origin on the scapula, while the other heads only functions as an elbow extensor (Williams *et al.* 2008b). This thesis focuses primarily on the lateral head as this is the part of the triceps brachii that is easiest to assess with the AMG-equipment. Next to the long head the lateral head is the biggest in triceps brachii with a weight of 129g in racing greyhounds and has an estimated maximum isometric force of 289N and a power of 9W (Williams *et al.* 2008b). For comparison, the long head weighs 341g and has a force of 1475N and a power of 58W. The maximum moment arm is 4.3cm at maximum extension of the elbow in greyhounds (Williams *et al.* 2008b). Triceps brachii, primarily the long head, is capable of producing extremely large amount of force and is the strongest muscle in both the thoracic and pelvic region (Williams *et al.* 2008b).

Picture 7 illustrate the movement of the limbs in a dog whilst walking. The purpose is to show when gluteus superficialis and triceps brachii are active in a dog walking according to approximate angles of the hip and elbow extensor, respectively. Triceps brachii stabilize the shoulder and elbow at touch down and is active as the elbow extends from touch down to the middle or late stance phase (Fischer & Lilje 2014; Cullen *et al.* 2017) (picture frames B-D).

In general triceps brachii play a major role in forelimb movement as an elbow extensor and can easily be assessed by the AMG-equipment.



Picture 5: Muscles of the forelimb from a dog, with triceps brachii high-lightened and the four heads described with origin, insertion and percentage of type I fibres. A: The muscles of the forelimb from a lateral aspect. B: The muscles of the forelimb from a medial aspect. (Armstrong *et al.* 1982; Miller 1993; Concha-Albornoz 2012; Fisher & Lilje 2014) The pictures are retrieved as screen-dumps June 22, 2017 from the video <https://www.youtube.com/watch?v=HSEN291vRAW> and modified.

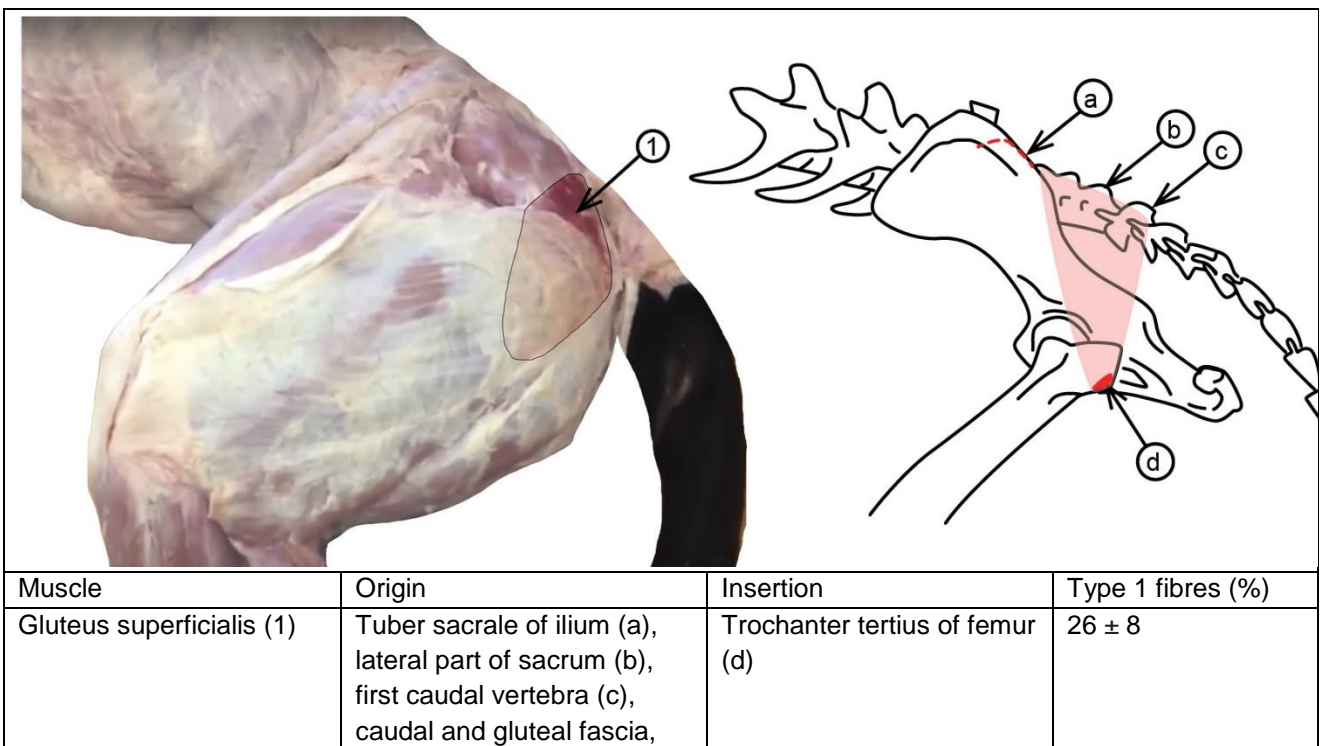


### 1.6.2 Gluteus superficialis

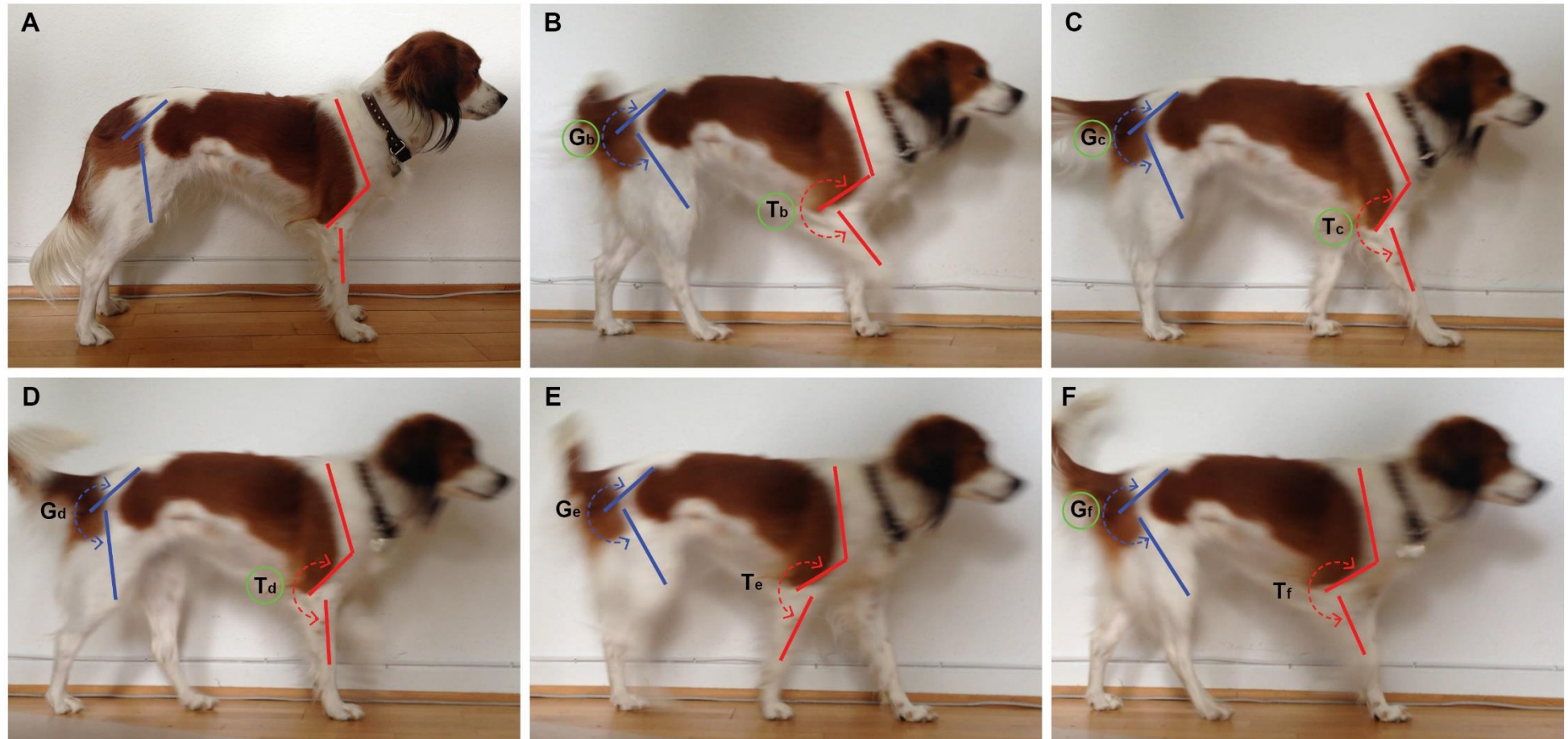
Gluteus superficialis are an extensor of the hip joint and innervated by the nerve gluteus caudalis (Miller 1993; Aspinnall & Cappello 2009; Fisher & Lilje 2014). It is positioned in the hip as illustrated in picture 6 and listed in the associated table. Gluteus superficialis cover a distal part of gluteus medius and in large dogs the muscle is 5 to 7 cm wide and more than 1 cm thick caudally (Miller 1993). It has a muscle mass of 158 g in racing greyhounds and an estimated maximum isometric force of 840N and a power of 17W (Williams *et al.* 2008a). The maximum moment arm is 0.3cm at maximum flexion of the hip 17W (Williams *et al.* 2008a). It has the highest ratio of muscle fascicle length : moment arm length of the hip extensors, indicating the ability of a muscle to move a joint through a large range 17W (Williams *et al.* 2008a).

Gluteus superficialis is active as the hip extends from late swing phase to the middle of the stance phase, and are partly responsible for propulsion of the body (Fischer & Lilje 2014; Cullen *et al.* 2017) (picture frames B,C,F).

In general, gluteus superficialis play a major role in hindlimb movement and propulsion work as a hip extensor and can easily be assessed by the AMG-equipment.



Picture 6: Muscles of the hindlimb from a dog with gluteus superficialis (1) high-lightened and skeleton showing the origin of gluteus superficialis (a,b,c) as well as insertion (d). The muscle is listed in a table with origin, insertion and percentage of type I fibres. (Armstrong *et al.* 1982; Miller 1993; Fisher & Lilje 2014) The picture is retrieved as screen-dumps June 22, 2017 from the video [https://www.youtube.com/watch?v=QuEL-8510fM&index=4&list=PLRRHuUzAO9qrY06\\_DetvI3Y0\\_OPO6dAWJ](https://www.youtube.com/watch?v=QuEL-8510fM&index=4&list=PLRRHuUzAO9qrY06_DetvI3Y0_OPO6dAWJ) and modified. The drawing is made by the author of this thesis with inspiration from Aspinnall & Capello (2009) and Miller (1993).



Picture 7: Visualization of difference of angles in hip (blue lines) and elbow-shoulder region (red lines) in a walking dog. Picture A shows the dog standing in a neutral position, while picture B-F shows the movement of a cycle when walking at a slow pace. The two blue lines mark the approximate distance from the iliac crest to ischial tuberosity and from the greater trochanter of the femur to the lateral femoral condyle respectively. The three red lines mark the approximate distance from the dorsal end of the spine in scapula to the greater tubercle of the humerus and further to the olecranon and the direction of the radius is shown as well. The angle G is illustrated as a dashed blue arch to show how and when gluteus superficialis work as a hip extensor and the angle T is illustrated as a dashed red arch to show how and when triceps brachii works as an elbow extensor when walking. A green circle around the angle indicate that the muscle is active in the transition from the current picture to the next. Notice  $G_b > G_c > G_d < G_e < G_f = G_b$  and  $T_b > T_c > T_d > T_e < T_f < T_b$ . (Miller 1993; Fischer & Lilje 2014; Cullen *et al.* 2017)



## 1.7 Muscle activity during the exercises

### 1.7.1 The warm-up exercise

Warm-up has been defined as having the function of improving muscle dynamics so that it is less inclined to injury and prepare the athlete for demands of exercise (Wood *et al.* 2007).

A study performed in Ohio showed that 88% of the handlers warmed their dogs up before participating in an agility competition and 58% made stretching exercises on their dogs (Zachary *et al.* 2014). No such research has been made in Denmark, but from the author of this thesis experience the percentage of handlers warming up their dogs could be the same while the method of stretching the dogs is far less common. Although no English written studies have been made to investigate the effect of active warm-up exercises in dogs, recommendations have been made for racing greyhounds prescribing 5 to 10 minutes of brisk walking or jogging before racing (Blythe *et al.* 1994 cited in Steiss 2002). Contrary many studies have been made in humans and some for horses, which will be used as model for the dogs in this thesis.

The purpose of warm-up is to increase body temperature 0.5°C - 1 °C (Steiss 2002). In humans, an increased body temperature lead to improved muscle perfusion (Gerbino *et al.* 1996), increased flexibility (range of motion) (Tsolakis & Bogdanis 2012) and improves neuromuscular coordination as well as increase transmission speed of nerve impulses (Karvonen 1992). It is suggested to reduce risk of injury as warm muscles injure higher forces before injury in rabbits (Strickler *et al.* 1990), but this has not yet been found to be the case in dogs performing agility (Cullen *et al.* 2013a; Zachary *et al.* 2014). Warm muscles improve the ability of collagen and myotendinous junction (site of connection between muscle and tendon) to deform and thus strengthen, but depending on whether the cause of injury is strain or load a warm muscle might be prone to injury due to greater deformation (Malone *et al.* 1996). It is also shown to reduce energy turnover at high intensity exercises (Ferguson *et al.* 2002), but at sub-maximal levels warm-up exercises results in a lower energy turnover (Ferguson *et al.* 2002) and reduced ability to perform (Gregson *et al.* 2002). In general, there are contradicting results in the numerous effect of warm-up exercises, as the studies varies both in methods to warm-up and type of the following exercise as they have no clear or common definition of warm-up exercises (Gerbino *et al.* 1996; Ferguson *et al.* 2002; Gregson *et al.* 2002; Bishop 2003; McMillian *et al.* 2006; Tsolakis & Bogdanis 2012). It should be noted that warm-up exercises should not result in muscle fatigue as this would compromise the subsequent performance.

Warm-up exercises (medium-intensity: 10 minutes trotting or high-intensity: 5 minutes trotting plus 14 seconds interval of running) for standardbred horses before sprinting has been shown to

lead to a higher oxygen consumption, reduced oxygen deficit and hence a decreased rate of anaerobic ATP-provision. It also resulted in a decreased rate of muscle glycogenolysis and a decline in blood and muscle lactate accumulation. No benefit of a high-intensity warm-up compared to a medium intensity warm-up was found. (McCutcheon *et al.* 1999) Similar results have been found in humans (Houmard *et al.* 1991 cited in McCutcheon *et al.* 1999).

Warm-up exercises can be classified as passive and active. In passive exercises, the dog is passive and the muscles are warmed by external heating, an example could be the use of warm water, stretching or massage. Pre-exercise massage have been suggested to reduce exercise-induced muscle injury in sled dogs, but no significant difference was found when measuring the degree creatine phosphokinase release between a control and a massage group suggesting that pre-exercise massage do not reduce the severity of exercise-induced muscle damage (Huneycutt & Davis 2015). In humans, a short stretching protocol has not shown to change performance while a long stretching protocol after warm-up exercise has shown to decrease performance (Tsolakis & Bogdanis 2012). Another study compared dynamic and static-stretching warm-up and found dynamic warm-up to result in a better performance in all three tests, while static-stretching only improved performance in one of the three tests compared to no warm-up exercise (McMillian *et al.* 2006).

The active warm-up exercise can be divided into general and specific warm-up. The purpose with the general warm-up exercise is to loosen up the muscles, and can be done with brisk walking or trotting (Malone *et al.* 1996; Steiss 2002). The specific warm-up exercise is also called the neuromuscular warm-up and is implemented to target the specific movement and muscles needed in the performance, and can be less intense work of the same exercises (Steiss 2002; Malone *et al.* 1996). This could especially be useful for dogs performing agility (Steiss 2002).

Performing cavaletti can be used as a specific warm-up for dogs (Drum *et al.* 2015). A cavaletti is 4-10 poles placed appropriate distance apart determined by the dog's natural stride length which differ according to speed. The height of the poles should be just above the carpus of the dog. The purpose of the cavaletti is to increase elbow, stifle and hock flexion and challenge the proprioception, balance and coordination and is also used as a therapeutic exercise after neurologic impairment in dogs (Drum *et al.* 2015).

After reviewing scientific studies about the effect of passive and active warm-up methods Behm & Chaouachi (2011) generally recommends a submaximal intensity aerobic activity followed by large amplitude dynamic stretching and then completed with sport-specific dynamic activities for a better performance and minimize impairments. Wenger *et al.* (1996) recommends a warm-up of low intensity suggesting 60 % of maximum oxygen consumption or 70% of maximal heart rate for

less than 15 minutes. Likewise, Bishop (2003) recommend intensity on the level of 40-60% of maximum oxygen consumption to increase body temperature and limit high-energy phosphate depletion.

From these recommendations, a warm-up protocol for dogs performing agility can be planned.

### 1.7.2 Hurdle

The jumping sequence has been described in horses with five phases: approach, take-off, aerial phase, landing phase and departure (Powers & Harrison 1999). The purpose of the approach is to ensure correct stride length, speed, balance and angular momentum all leading to conditions for the take-off. In the take-off, the front limbs make a braking force to reduce the time the hindlimbs act to make the propulsion movement. The animal must obtain an optimum vertical velocity and rotation as well as angular momentum when being in the stance phase with the two hind limbs. As the hind limbs leaves the ground the animal enters the suspension phase in which the animal will undergo a vertical and horizontal displacement while controlling the rotational element of the movement. In the suspension phase the animal must prepare for the landing phase in which the front limbs firstly take the touchdown asymmetrically and ensure the hindlimbs stay clear of the fence. Meanwhile the impact of landing is absorbed while retaining sufficient horizontal velocity being ready for departure. The purpose of the departure is to make a smooth transition from landing to the approach of the next jumping sequence. (Powers and Harrison 1999)

Gear ratio<sup>1</sup> at the limb joints (wrist, elbow, shoulder, ankle, knee and hip) has been investigated in three large dogs (mean=20kg) jumping vertically with maximum performance to the top of an elevated platform (0.845-0.905cm high) (Gregersen & Carrier 2004). They found a drop in the extensor muscle gear ratio in the wrist, elbow and ankle joints which is suggested to improve the potential for storage and recovery of elastic strain energy. Meanwhile the capacity of storing and recover elastic energy is not found in the hip joint when jumping as the hip joint only extends during jump.

The kinetics of jumping in agility dogs have been investigated (Pfau *et al.* 2010; Birch & Lesniak 2013; Birch *et al.* 2015). Pfau *et al.* (2010) investigated the effect of distance between hurdles and the difference of 1.5-meter-long jump and 0.6-meter-high hurdles when landing using 11 border collies with agility experience. They found that dogs jumping a 0,6m hurdle had a higher peak vertical force than dogs jumping a long jump, meaning they land with more force after the jump (N/kg BM), implying a higher workload on the body and muscles. They also had a steeper landing

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<sup>1</sup> Gear ratio for a muscle is ground reaction force moment arm (computed) divided with muscle moment arm (measured on the dog). See figure 1 in Gregersen & Carrier (2004).

after a high jump. The highest vertical force (45 N/kg BM) was found on forelimb pair after a hurdle jump with 5 meters' distance and at high speed. More weight was put on the front limbs with a weight distribution of 60%/40% when jumping hurdles. The decelerative force was higher on the forelimbs compared to hind limbs when landing from a hurdle and long jump. They had a decreased stance time during hurdle jumps indicating that dogs could not distribute the forces over a longer time period. Finally, they also found that lower jumps resulted in higher speed when comparing long jump and hurdles.

The forelimbs are loaded asymmetrically when landing (Pfau *et al.* 2010; Meershoek *et al.* 2001). The trailing limb endure a higher vertical force and is placed first when jumping hurdles compared to the leading limb and (Meershoek *et al.* 2001; Pfau *et al.* 2010). They each do different work according to decelerating or accelerating work in the landing phase. In horses, the leading limb make decelerative work in the first part of the stance phase while the trailing limb make accelerative work during the whole stance phase (Meershoek *et al.* 2001). Meanwhile with increasing hurdle height the accelerative force increases in dogs due to the requirement of converting potential energy into forwards kinetic energy (Pfau *et al.* 2010).

Birch & Lesniak (2013) investigated the effect of hurdle height on joint angles on 8 agility dogs. The height of the hurdles was set as a percentage according to the height at the withers of the dogs, with 7 % lower as the lowest and 51 % higher as the highest. The angle of the tarsus and sacroiliac region had a significant increase in extension during take-off with increased hurdle height. Increased flexion was found for the radio-humeral and scapula-humeral joints and the base of the neck at take-off with increased hurdle height. No significant difference according to joint angles was found in landing, although landing distance increased significantly with increased hurdle height, as oppose to the take-off distance that was unaltered.

Birch *et al.* (2015) investigated the effect of altered distances (3.6, 4.0 and 5.0 meters) between obstacles in 54 large agility dogs. The dogs had a significantly closer take-off and landing distance and a lower speed in the following order: 4 m distance was lower than 3.6 m distance and both were lower than 5 m distance. The only deviation from this was the distance in landing for 3.6 m and 4 m in which there were no significant difference.

Again in 2016 Birch *et al.* investigated how jump kinematics altered as hurdle height increased. They analysed the jumping trajectory of 19 border collie and a border collie cross jumping three hurdles with five meters distance in heights from 0 to 15 to 65 cm with an increased height of 10 cm between each trial. The percentage height (percentage of the hurdle height in relation to the dogs' height at the withers) was determined and categorized to compare the results and for further analysis. They found a significant decrease in speed, more acute neck angle and more

flexed shoulder angle when jumping > 76% of their height to the withers. The length of trajectory significantly increased when dogs jumped between 51% - 125% of their height while decreasing when jumping >126% of their height, but still neck angle became more acute. Lumbar spine angles became significantly more extended dorsally when jumping > 101% of their height.

With the use of fine wire electromyography (see section 1.8 *Approaches for measuring of muscles*) the activation of four forelimb muscles (biceps brachii (BB), supraspinatus (SP), infraspinatus (IF) and triceps brachii – long head (TBLH)) was assessed in eight border collies jumping two 55cm high hurdle positioned 4,5 meters apart (Cullen *et al.* 2017). The jumping sequence of the hurdle was assessed in three strides: the pre-transition stride (stride immediately before jumping), transition stride (from legs are lifted off the ground until landing) and post-transition stride (stride immediately after jumping). The results were compared to baseline-measurements of the dog when walking. The peak activation across all four muscles was higher when jumping compared to walking and peaked in the swing phase, except for the transition strides which had two peaks, one at early stance and one during mid-swing. The highest peak activation for TBLH was found in the swing phase during transition for the first jump with 10.6 times walking. Visually several dogs had a higher peak activation at first jump compared to the second, but no significant difference was found, this could be due to a small sampling size. Likewise, there seemed to be a difference in peak muscle activity when the number of pre-strides were taken into account, but there were not enough data to run statistical analysis. The same study investigated the activation in the four forelimbs when running the A-frame.

### 1.7.3 A-frame

As with the jumping-sequence the performance of an A-frame may also be divided in 5 phases, approach, ascending, aerial phase (above apex), descending and departure. In this trial, the ascending and descending phase is of interest and described in section 2.7.3 *The A-frame*.

Walking at an incline is an isotonic concentric exercise, while walking at a decline is an isotonic eccentric exercise (Lauer *et al.* 2009). Both occurs when running an A-frame. Ascending an A-frame is an isotonic concentric exercise, while descending an A-frame is an isotonic eccentric exercise.

#### *Forelimb*

Besides measuring the activation of four forelimb muscles in eight border collies jumping hurdles Cullen *et al.* (2017) also assessed the effect of running the A-frame set in two heights: 1.67 and 1.75 meters at apex. The running sequence of ascending and descending the A-frame was each assessed in three strides: the pre-transition stride (stride immediately before transition stride), transition stride (ascending: from legs are lifted off the ground until paw down on the A-frame,

descending: from legs are lifted off the A-frame until landing on the ground) and post-transition stride (stride immediately after transition stride). The results were compared to baseline-measurements of the dog when walking. As with the hurdles the peak activation across all four muscles was higher when running the A-frame compared to walking. Of the four muscles, the TBLH had the lowest peak activation descending the A-frame at pre-transition stride regardless of the height, and the low peak activation continued in the transition when descending the A-frame until the post transition stride. In contrast TBLH had some of the highest peak activations when ascending the A-frame. The activation of the four muscles peaked in the swing phase of the gait cycle when ascending the A-frame, but when descending the A-frame the peak activation occurred in stance for many strides. Also, when descending the A-frame a greater proportion of the stride was spent in stance. No significant difference was found in peak activation across all four muscles comparing the high and the low A-frame.

Carrier *et al.* (2008) found that the protractor and retractor muscles of the forelimb (pectoralis superficialis descendens, pectoralis profundus (posterior portion), omotransversarius and thoracic portion of the trapezius) had an increased activity ranging from 1.9 to 4.2 times when trotting 14° uphill, compared to when trotting at normal level. Likewise, some muscles of the forelimb (pectoralis superficialis descendens and omotransversarius) had an increased activity ranging from 2.0 to 2.5 times when trotting 14° downhill, compared to when trotting at normal level. Other forelimb muscles (pectoralis profundus (posterior portion) and latissimus dorsi) had a significant decrease in activity ranging from -0.4 to -0.6 when trotting 14° downhill, compared to when trotting at normal level. Thus, the propulsive work when running uphill require an increased activity in most forelimb muscles investigated, while the breaking work downhill resulted in both an increased and decreased activity on different forelimb muscles. The duration the muscles was active did also increase in several muscles when running uphill, but were less pronounced when running downhill. Some muscles (cleidobrachialis) had so little activity at normal level that the activity when running up- and downhill increased several hundred times, but due to variation it was not statistical significant.

### *Hindlimb*

Lauer *et al.* (2009) investigated the effect of 5% incline and decline on muscle activity and joint range of motion in hind limbs of eight dogs with the use of EMG. Although range of motion in the maximal stifle joint flexion and hip joint decreased when going from an 5% incline to a 5% decline, the muscle activity of gluteus superficialis were unchanged comparing 5%, 0% and -5% inclination.

In horses, the muscle activity of gluteus medius are increasing when the slope increases up to 6% as a result of the amount of energy required to raise the body upward and increased time per

stride. The major function of the gluteus medius when trotting uphill is the propulsion of bodyweight during the stance phase (Robert *et al.* 2000), and the same could be the case for gluteus superficialis. Robert *et al.* (2000) suggest that a higher electromyographic activity, found in gluteus medius in horses running uphill (6%), require recruitment of additional motor units.

### *Contact method*

No previous studies have been made to investigate how the different contact-method practised on the A-frame affect the muscles of the dog. As the 2o2o-contact method require a full stop the muscles could be subjected to a large breaking force, both when standing in the 2o2o position, but also doing decelerative work on the run down the A-frame (Carrier *et al.* 2008; Cullen *et al.* 2017). Additionally, more weight would be loaded on the front limbs as the dogs stand with the hindlimb uphill. In contrary, when practicing the running A-frame a requirement to convert potential energy to forwards kinetic energy could be expected and increased with increased height of the A-frame as seen when jumping higher hurdles (Pfau *et al.* 2011).

### 1.7.4 The cool-down exercise

In Ohio 57% of the handlers cooled their dogs down after participating in an agility competition and 25% made stretching exercises on their dogs (Zachary *et al.* 2014). No such research has been made in Denmark, but from the author of this thesis experience the percentage of handlers stretching their dogs would likely be lower.

While only few studies have been made on the effect of warm-up in dogs nothing is made for dogs performing post-event exercises or cool-down exercises and few in humans and horses as well. During cool-down exercise the blood circulation and pulse gradually decreases, and working muscles can remove lactic acid more efficiently than resting muscles (Krzywanek 1988). It is generally recommended to continue with a low-intensity-exercise after performing to wash out the accumulated lactic acid and shorten recovery time in the muscles (Baltzer 2012). The optimal level is found to be trotting about 350-450 m/min for horses (Krzywanek 1988) while in dogs 10-20 minutes of easy trot or walking (35-65% of maximal oxygen consumption) is recommended (Baltzer 2012), but massage or stretching could also be applied (Herbert & Gabriel 2002; Larsen 2013). For comparison, in humans, the maximum rate of removal of lactic acids is found when working 40% of the maximum oxygen uptake (Davies *et al.* 1969). When the oxygen supply in the cells are insufficient pyruvate and hydrogen is combined to produce lactic acid, which is a good energy source (Robert *et al.* 2004). Lactic acid has falsely been accused of causing acidosis and muscle fatigue, while the opposite is the case (Robert *et al.* 2004). There is however an increased proton release under intense exercises, but this is due to the production of ATP from glycolysis and causes acidosis in the muscles. This is why it is recommended to make cool-down exercises



after a performance, and although the performance of cool-down exercises is said to decrease risk of injuries it is not proven to do so (Herbert & Gabriel 2002; Cullen *et al.* 2013a).

Six horses performing 2 times 10 minutes of trot and 3 minutes of gallop had a decreased ESTi-score in gluteus superficialis and semitendinosus as well as an imbalance in semitendinosus, when compared with an initial walk (Harrison 2017b). After 10 minutes of a post-exercise walk the balance and ESTi-scores reached same level as during the initial walk, but they did not compare measurements of walking initially and finally in the post exercise walk.

The cool-down exercise is implemented to investigate if cool-down exercises has any immediate effect on the muscles.

## 1.8 Approaches for measuring of muscles

Different approaches and equipments have been developed to measure muscle activity and condition in dogs. Electromyography (EMG) is a method to assess muscle function by measuring the electric potential difference between two electrodes and thus measure the work of the motor units (Pullman *et al.* 2000). EMG can be used both invasive as needle electromyographic (nEMG) or fine-wire EMG (fEMG) or noninvasive as surface EMG (sEMG) (Pullman *et al.* 2000). The latter is the most often used opposed to nEMG and fEMG which is both invasive and painful (Pullman *et al.* 2000). The problem with sEMG is that the measurements are affected by both technical, experimental, descriptive as well as physiological circumstances as summarized by Harrison (2017a). The positioning of the sensor is intricately and only small differences in the location may result in a large variability in the recorded signals (de Luca 1997, Harrison 2017a). Altered position of electrodes, as the muscle fibre length changes during locomotion, may also alter the sEMG signal (Lauer *et al.* 2009). The sensors must be close to the skin and no moist must come in-between, because the skin may become shunted and the signal amplitude decreases (De Luca 1997). For the sensors to be close to the skin the skin must be exposed by shaving and thoroughly cleaned (Lauer *et al.* 2009). The duration of an exercise effects the extracellular fluids which may affect the sEMG recordings and thus confound the activation of motor units (Harrison 2017a). Surface EMG cannot be used to distinguish between concentric and eccentric contractions (Harrison 2017a). Fine-wire EMG have been used to measure the magnitude of muscular activation of four canine forelimb muscles in eight dogs performing a two-jump sequence and running an A-frame at two different heights (Cullen *et al.* 2017). Using this invasive method trimming of hair and anaesthetic for the skin was required as well as a real-time ultrasonography was needed to guide the needle insertion of the fine wire electrodes which had to be done by certified veterinary radiologists (Cullen *et al.* 2017)



Multi-frequency bioimpedance (BIA) is an alternative non-invasive assessment of the muscles and besides being used for humans (Bartels *et al.* 2015) it has proved to be useful in diagnostic and training of horses (Harrison *et al.* 2015) as well as to evaluate information of muscle health in dogs (Fenger 2017). BIA measures impedance at several frequencies and thus analyses the relative volume of fluid compartments resulting in information about both extracellular and intracellular conditions of the muscle (Ivorra 2002, Bartels *et al.* 2015). The BIA-equipment can only be used while the animal stand completely still and is free of metal objects (Harrison *et al.* 2015). Dogs need to be shaved for the electrodes to reach the skin which, if incautious, may result in irritated skin or inflammations (Fenger 2017). The method will only provide a still picture of the muscle condition and cannot show a real-time effect of an active dog. While BIA measure static muscle conditions another method is developed to assess dynamic muscle conditions.

### 1.8.1 Acoustic Myography

Acoustic Myography is a method to assess the muscles measuring muscle-sound (Harrison *et al.* 2013). The AMG-technique transdermally measures the pressure waves that are generated by the vibrations when muscle fibres contract (Bartels *et al.* 2017, Fenger & Harrison 2017a). An AMG system (CURO) has been developed to measure three parameters as listed and described in table 2, 3 and 4. With the use of a contact transducer with piezoelectric crystals, it has become possible to use AMG for more accurate as well as repeatable muscle sound recordings (Harrison *et al.* 2013). The new wireless CURO (MyoDynamik ApS, Frederiksberg, Denmark) was evaluated in 2013 by Harrison *et al.* and found to be able to determine coordination as well as aspects of muscle function in physically active and healthy humans. The CURO has since been applied to both human (Harrison *et al.* 2013, Harrison 2017a, Bartels *et al.* 2017), horses (MyoDynamik ApS 2017, Riis *et al.* 2013) and dogs (Fenger & Harrison 2017a). No specialist technical knowledge is needed for the person carrying out the recordings (Harrison *et al.* 2013) and is, together with being wireless and a non-invasive technique, easy to apply in multiple situations. It has been used to study humans running in the city (Harrison *et al.* 2013), lameness in a horse (Riis *et al.* 2013), comparison of trot and walk in dogs (Fenger & Harrison 2017a), and latest the effect of 10 months of training in a BMX rider (Bartels *et al.* 2017), thus shown to be efficient when measuring active subjects.

### 1.8.2 The AMG-equipment

The AMG-equipment comprises of: A CURO, sensors with wires, Snøgg, EKO GEL, a canine harness from Julius-K9®, the app CURO Equine installed on an Ipad and an online program.

The CURO, see picture 8, has been developed by the Danish company MyoDynamik ApS, for the purpose of measuring AMG. The CURO collect signals via unidirectional sensors with piezoelectric crystals covered by a sounding board with a diameter of 4 cm positioned above the muscle of interest (Harrison *et al.* 2013). The wire which connect the sensors to the CURO is 2 meters of length. When turned on the CURO record and store all data for later analysis in the online program [curo.softtheme.com](http://curo.softtheme.com) also developed by MyoDynamik ApS. Additionally, the CURO is able to send a signal in a 100-meter distance via Wi-Fi to an Ipad with the app CURO Equine installed. CURO Equine can be installed from the App Store for free, but it only works on Ipad with iOS 8.0 or newer versions. CURO Equine makes it possible to see real-time measurements of the muscles visualising the soundwaves and giving both balance and ESTi™-parameters, with the purpose of on-location-interpretation and to ensure the sensors functions correctly. An acoustic gel can be added to reduce impedance between skin and sensor and ensure a good connection and exclusion of extraneous noise (Harrison *et al.* 2013, Harrison 2017b,b). The use of Ultra-Sound Gel (BlueScan – Lina Medical ApS, Glostrup, Denmark) may simultaneously lower the frequency range to ~5 Hz (Harrison *et al.* 2013) alternatively EKO GEL (Ekkomarine Medico A/S, Holstebro, Denmark) can be applied, see picture 9. The sensors must be fastened to the subject without restriction of movement. This can be done with the use of a self-adhesive bandage such as Co-Flex (Co-Flex, Andover - Salisbury, MA) (Harrison *et al.* 2013) or Snøgg (Snøgg AS, Kristiansand, Norway), see picture 10. The CURO must also be fastened to the subject, and for dogs it fits perfectly under the handle on a canine harness from Julius-K9®, (Julius-K9®, Szigetszentmiklós, Hungary) (Fenger & Harrison 2017a), see picture 11 and additionally picture 16 in section 2.6.3 *The mounting of AMG-equipment.*



Picture 8: The CURO and sensors used to measure muscle sound. The picture is retrieved August 7, 2017 from <http://www.myodynamikequine.com/amg/>



Picture 9: Acoustic gel (Ekkomarine Medico A/S, Holstebro, Denmark) used to remove impedance at the surface between sensor and skin.



Picture 10: Snøgg (Snøgg AS, Kristiansand, Norway) used to fasten sensors to the skin of dogs.



Picture 11: Canine harness (Julius-K9®, Szigetszentmiklós, Hungary) used to fasten the CURO without restriction of movement.

### 1.8.3 Interpretation of the ESTi™-scores

The collected data in the form of sound signals are analysed and transformed to an ESTi™-score. The ESTi™-score consist of three parameters E-, S- and T-parameter and the average of these scores combined is an ESTi-score. Each parameter is given a score from 0-10, 0 being the poor condition and 10 being the perfect condition of the muscle, but scores above 7-8 is considered optimal. A low ESTi™-score would be interpreted as the subject having a poor coordination (E), many fibres recruited (S) with a high firing rate (T) Meanwhile a high ESTi™-score would be interpreted as the subject having a good coordination (E), few muscle fibres recruited (S) with a low firing rate (T). This is further explained and illustrated in tables 2,3 and 4. Each score is given in relation to a maximum value administered in the program on [curo.softtheme.com](http://curo.softtheme.com). (MyoDynamik ApS 2017)

When a muscle meets the need for an increased force the central nervous system may either increase the number of muscle fibre recruited or increase the firing rate, resulting in a decreased S- or T-score respectively (MyoDynamik ApS 2017). An improved use of the muscle can be seen with an increased E-score and this will also be reflected in the T- and S-score (Bartels *et al.* 2017).

Muscle fatigue can be revealed with the use of a CURO. An early sign of muscle fatigue is a temporary increase in recruitment of fibres or/and a higher firing rate, as a short burst before the muscle fatigue and the recruitment of fibres and firing rate of muscle fibres decrease rapidly. Imbalance in the use of muscles on each side of the dog will also be more pronounced when the muscles are fatigued. Muscle fatigue is also seen by a decreasing E-score due to an increased

active period compared to inactive period of motor neurons, as a result of the muscles being less coordinated. An improvement is seen when the opposite happens as the muscle become more coordinated. (MyoDynamik ApS 2017)

Generally, low E-scores and the combined ESTi-scores tell the muscle cannot cope with the amount of work inflicted on it and it may be in risk of injury due to overload.

Table 2: Explanation and interpretation of the E-parameter gained from acoustic myography signal using a CURO. General interpretation of the parameters as well as high and low scores is explained with illustrating pictures.

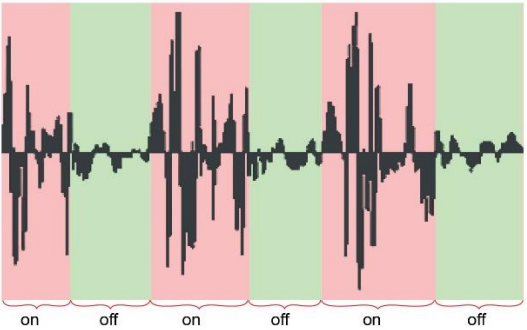
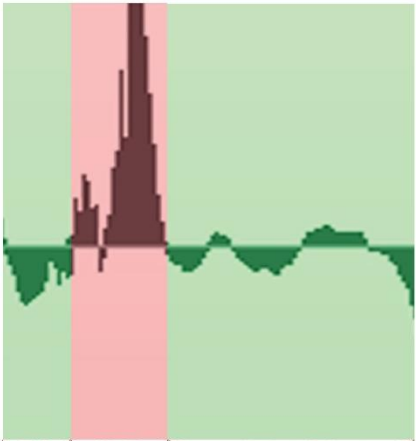
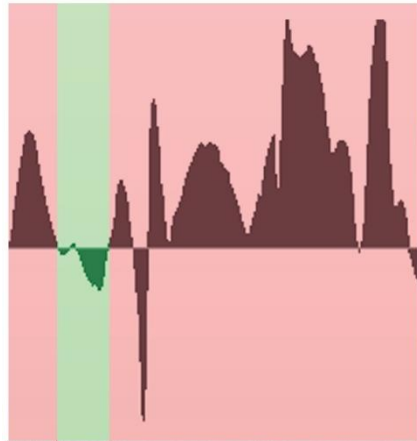
	Interpretation of the parameters	High score	Low score
<p>E-parameter</p>	<p>Coordination and efficiency of the muscle. The E-parameter informs how synchronized the motor neurons is activated, by measuring the duration of inactive and active periods compared to active periods. (MyoDynamik ApS 2017; Harrison 2017a)</p>  <p>1.18 seconds</p> <p>Sound signal from triceps on a Tervueren trotting after 15 minutes of cool-down exercises. X-axis is time elapsed and y-axis is amplitude. Red markings with the label "on" indicates active periods in which the motor neurons are turned on and the green markings with the label "off" indicate the inactive periods in which the motor neurons are turned off. E-score = 3.</p>	<p>A high E-score shows a good coordination/efficiency of the muscle. The activation of the motor neurons is perfectly synchronized. This will result in a narrow active period. This is seen in well trained animals. (MyoDynamik ApS 2017)</p>  <p>0.22 seconds</p> <p>Sound signal from triceps on a Tervueren trotting after 15 minutes of warm-up exercises. X-axis is time elapsed and y-axis is amplitude. Red markings indicate active periods while green markings indicate the inactive periods. E-score = 5.</p>	<p>A low E-score shows a poor coordination/efficiency of the muscle. The activation of the motor neurons is not synchronized. This will result in a broader active period. This is seen in poorly trained animals. (MyoDynamik ApS 2017)</p>  <p>0.22 seconds</p> <p>Sound signal from triceps on a Tervueren trotting before warm-up exercises. X-axis is time elapsed and y-axis is amplitude. Red markings indicate active periods while green markings indicate the inactive periods. E-score = 2.</p>

Table 3: Explanation and interpretation of the S-parameter gained from acoustic myography signal using a CURO. General interpretation of the parameters as well as high and low scores is explained with illustrating pictures.

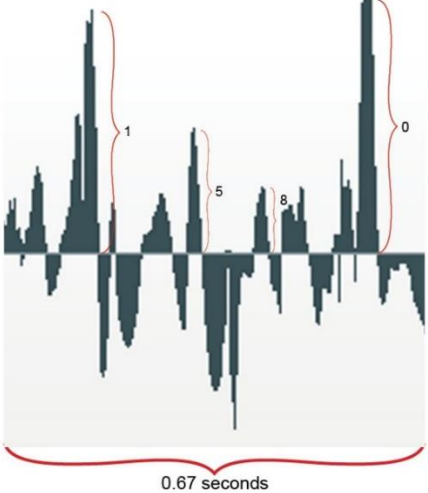
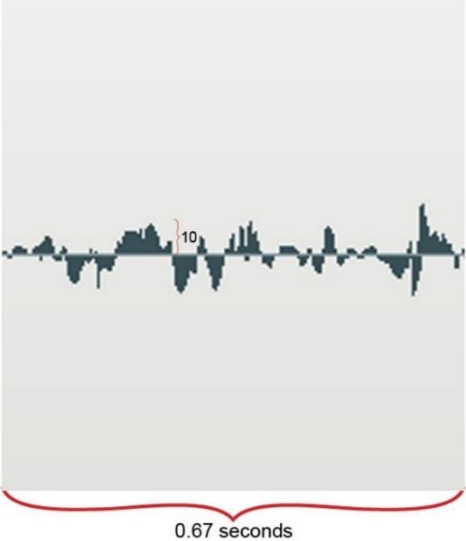
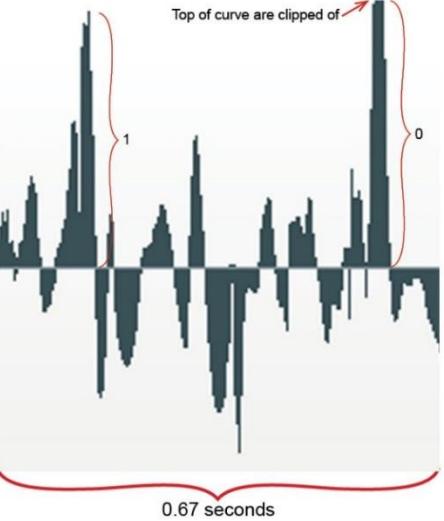
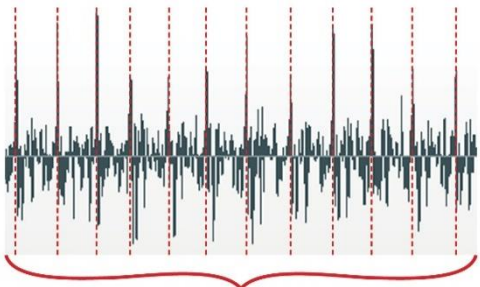
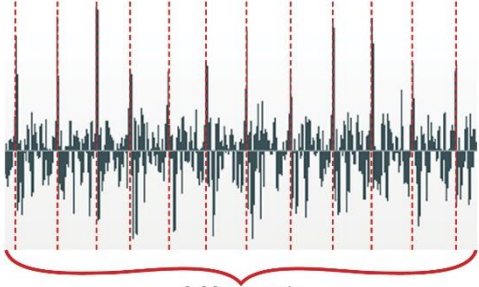
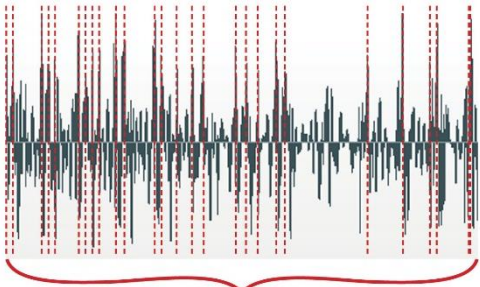
	Interpretation of the parameters	High score	Low score
S-parameter	<p>Quantity of active fibres recruited. The S-parameter informs how many motor units are recruited and synchronized as each motor neurons being activated has an additive effect on the force being produced (spatial summation) and can be seen as the amplitude. (MyoDynamik ApS 2017; Harrison <i>et al.</i> 2013)</p>  <p>0.67 seconds</p> <p>Sound signal amplitude from triceps on a Tervueren trotting after 15 minutes of warm-up exercises, showing different S-scores. X-axis is time elapsed and y-axis is amplitude.</p>	<p>A high S-score is a sign of few fibres being recruited. This will result in a low amplitude. (MyoDynamik ApS 2017)</p>  <p>0.67 seconds</p> <p>Sound signal amplitude from gluteus superficialis on a Tervueren trotting after 15 minutes of warm-up exercises, showing a high S-score. X-axis is time elapsed and y-axis is amplitude.</p>	<p>A low S-score is a sign of many fibres being recruited. This will result in a high amplitude. (MyoDynamik ApS 2017). If the amplitude exceeds the maximum range of 6db the S-score = 0 is assigned.</p>  <p>0.67 seconds</p> <p>Sound signal amplitude from triceps on a Tervueren trotting after 15 minutes of warm-up exercises, showing high S-scores and an amplitude being clipped of due to the sound exceeding the maximum range of recordings for the sensors. X-axis is time elapsed and y-axis is amplitude.</p>



Table 4: Explanation and interpretation of the T- and ESTi-parameter gained from acoustic myography signal using a CURO. General interpretation of the parameters as well as high and low scores is explained with illustrating pictures.

	Interpretation of the parameters	High score	Low score
T-parameter	<p>The firing rate of the active fibres. The T-parameter informs the frequency of activation of motor neurons (temporal summation), and can be seen as the sound signal frequency. (MyoDynamik ApS 2017; Harrison <i>et al.</i> 2017)</p>  <p>6.00 seconds</p> <p>Sound signal frequency from triceps brachii on a mixed breed walking after 10 minutes of warm-up exercises, showing each cycle with a dotted red line. X-axis is time elapsed and y-axis is amplitude.</p>	<p>A high T-score is a sign of a low firing rate of the active fibres. This will result in a low frequency. (MyoDynamik ApS 2017)</p>  <p>6.00 seconds</p> <p>Sound signal frequency from triceps brachii on a mixed breed walking after 10 minutes of warm-up exercises, showing a high T-score = 9. X-axis is time elapsed and y-axis is amplitude.</p>	<p>A low T-score is a sign of a high firing rate of the active fibres. This will result in a high frequency. (MyoDynamik ApS 2017) If the frequency exceeds the maximum range of 120hz the T-score = 0 is assigned</p>  <p>6.00 seconds</p> <p>Sound signal frequency from triceps brachii on a mixed breed trotting after 2 minutes of warm-up exercises, showing a low T-score = 6. X-axis is time elapsed and y-axis is amplitude.</p>
ESTi-parameter	<p>The overall performance of the muscle fibres. The ESTi-parameter give an overall insight to the status of the muscle, and is the average of the E-, S- and T-score combined.</p>	<p>A high ESTi-score is a sign of an overall well-trained and coordinated muscle with few muscle fibres recruited and a low firing rate.</p>	<p>A low ESTi-score is a sign of an overall poorly-coordinated muscle with many muscle fibres recruited and a high firing rate.</p>

## 2.0 Materials and methods

### 2.1 Recruitment of participants

Participants for the experiment were found through the social media Facebook making postings (Appendix B) on the Facebook pages “Agilitysiden DK”, “Border Collie Klubben” and “Border-Collie Danmark”. At first the post only targeted dog owners participating in agility with the breed border collie, for more equal measurements, minimizing the variables and making it easier to see statistical difference. Border collies is the most dominant breed in agility (Cullen *et al.* 2013a,b; Levy *et al.* 2009; Zachary *et al.* 2014) and thus it were more likely to find participants with this breed. As there came no response to the post, all owners with a dog participating in agility in the highest class according to the FCI regulation were targeted. This was done to set a minimum limit for the size of the dogs participating. The reason was both to ensure that the dogs were capable of, and used to jumping the high hurdles, and the dogs were capable of running and jumping with the weight of the CURO (300g) on their back. Only healthy dogs above 1.5 years of age was allowed to participate.

Participants were also found through private contacts from training or competition and some participated with two or three dogs in the experiment. All contact with the participants was in Danish.

As a thank for their time each participant was offered a bag with treats, dogfood and a water bottle sponsored by Royal Canin Nordics, Copenhagen S, Denmark.

### 2.2 Descriptive data of the experimental dogs

Ten dog owners with a total of 17 dogs participating in agility was assigned to the experiment, meanwhile 3 dogs were excluded due to non-agility-related injuries. One handler was injured during the trials due to a bad knee and the author of this thesis ran with the dog (Stella) instead.

Data was collected for 14 dogs participating in the experiments as listed in table 5 and continued in table 6.



Table 5: Descriptive data of the 14 experimental dogs, including name of the dog, breed, gender, if the dog is intact or castrated/neutralized, height to the withers, weight and age. For the data with numeric values the mean, median, minimum and maximum are listed.

Dog	Breed	Gender	Intact	Height (cm)	Weight (kg)	Age (year)
Misha	Shetland sheepdog	Male	no	46	13	3.0
My	Border collie	Bitch	yes	49	18	3.0
Jamie	Border collie	Male	yes	52	18	1.5
Jet	Border collie	Bitch	yes	55	18	8.0
Inga	Tervueren	Bitch	yes	59	18	3.0
Viola	Tervueren	Bitch	yes	56	20	4.5
Jubi	Tervueren	Bitch	yes	62	22	6.5
Tara	Australian shepherd	Bitch	yes	50	18	2.0
Ludvig	Australian shepherd	Male	yes	57	25	8.0
Diego	Golden retriever	Male	yes	55	28	4.5
Daphne	Old English sheepdog	Bitch	no	61	36	8.0
Stella	Labradoodle	Bitch	no	64	29	4.5
Wess	Mixed Breed	Male	no	54	23	2.5
Thor	Mixed Breed	Male	no	63	30	2.5
Mean	-	-	-	55,9	22,6	4,4
Median	-	-	-	55,5	21,0	3,8
Minimum	-	-	-	46	13	1,5
Maximum	-	-	-	64	36	8,0

Table 6: Descriptive data of the 14 experimental dogs, including name of the dog, years of experience in agility, body condition score on a scale from 1-10, score of how athletic the dog is built on a scale from 1-5, the training level measured in how many hours a week the dog practice, the height of the hurdle in normal training situation and the practice of contact method when running the A-frame being either the 2on-2off contact method (2o2o) or running contact method (run). For the data with numeric values the mean, median, minimum and maximum are listed.

Dog	Body condition score (1-10)	Athletic score (1-5)	Experience (Year)	Training level (h/week)	Hurdle height (cm)	Contact method
Misha	5	1	2.0	2-3	55	2o2o
My	5	1	2.5	<1	55	2o2o
Jamie	4	1	1.0	2-3	55	2o2o
Jet	4	1	7.0	<1	65	2o2o
Inga	5	2	2.0	<1	55	run
Viola	4	2	3.0	<1	65	2o2o
Jubi	5	2	2.0	<1	55	2o2o
Tara	4	1	1.0	1-2	45	2o2o
Ludvig	4	2	7.5	<1	55	2o2o
Diego	5	4	2.0	1-2	50	2o2o
Daphne	5	4	6.0	1-2	55	run
Stella	5	2	0.5	<1	45	run
Wess	4	2	1.5	1-2	45	2o2o
Thor	4	2	0.2	<1	55	2o2o
Mean	4,5	1,9	2,7	-	53,9	-
Median	4,5	2,0	2,0	-	55,0	-
Minimum	4	1	0,2	<1	45	-
Maximum	5	4	7,5	2-3	65	-

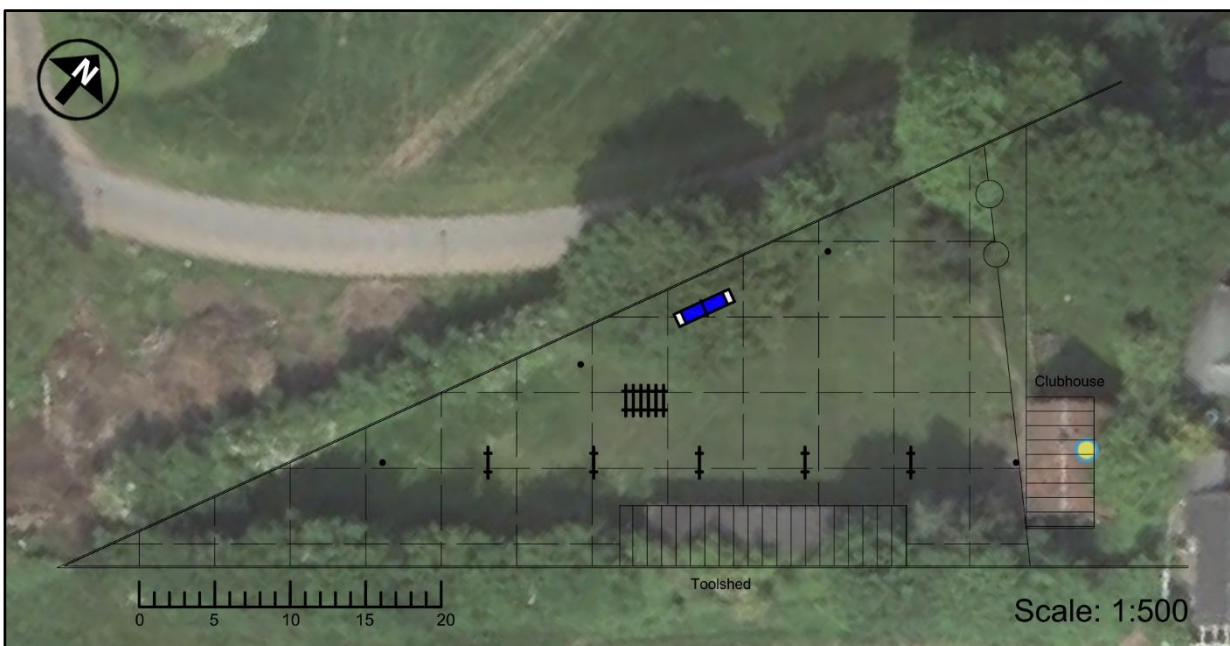
## 2.3 Pilot study

The AMG-equipment and the protocol was tested on a large Shetland sheepdog (height: 46 cm, weight: 13 kg), followed by corrections to the protocol and tested one more time following these corrections. In the first test trapezius was considered together with gluteus superficialis and the measurements looked perfectly fine. Due to concerns about the sensors position close to the harness and risk of interference with muscle sound when the harness moved above the sensor the trapezius was avoided and triceps brachii was chosen. The participants were sent an e-mail (appendix C) with practical information and possible dates to participate. Attached to the mail was a map with a description of the location of the training area and parking opportunities, a short description of the project and also the protocol (appendix D, E and F). The practical information contained a notification that the dog should not be warmed up, but that it was allowed to walk from the parking lot to the training area.

After receiving a reply with possible dates from the participants they were each administered a date and time between October 27<sup>th</sup> to November 16<sup>th</sup> 2016.

## 2.4 The location

The experiment took place at the training area belonging to The Danish Agility Association Copenhagen (Dansk Agility Forening København), dakbh@gmail.com, www.agility-kbh.dk at the address Lersøstien 175, 2100 Copenhagen East. Eleven hurdles, 4 poles and an A-frame were placed as illustrated on picture 12, 13, 14 and 15.



Picture 12: Position of the obstacles used for the experiment at the location Lersøstien 175, 2100 Copenhagen East. Five hurdles are positioned furthest south-east with a distance of 7 meters and with a pole in each end. Further north-west 6 hurdles are positioned with a distance of 0.5 meters. The A-frame is positioned along the west-northern fence with a pole 7 meters from each end. The drawing is in scale 1:500.

## 2.5 Exercise equipment

All the agility obstacles follow the regulations set by the FCI and was borrowed from The Danish Agility Association Copenhagen. A-frame equivalent to the obstacle used can be bought at Dong-Agility, Kim and Ann-Britt Gravlund-Krat, Dongsvej 9, 5854 Gislev, tel. 2078 4760. Likewise, hurdles can be bought at Hundebiksen Hans Peter Dinesen, Farup Grønnevej 4, 6760 Ribe, tel. 7542 1979.

### 2.5.1 The cavaletti

The dogs performed a cavaletti-exercise as a part of the warm-up protocol. The cavaletti composed of 6 hurdles positioned with a distance of 50 cm from centre to centre of the bar and a height of 20 cm. This is also illustrated in picture 13. The set up was the same for all dogs no matter the size of the dogs.



Picture 13: Set up of cavaletti with 6 hurdles in a distance of 50 cm from centre to centre and a height of 20 cm.

### 2.5.2 The hurdles

As seen in picture 14, 5 hurdles were placed in front of each other with a distance of 7 meters. As the effect of distance between hurdles are of no interest, the maximum distance recommended by the FCI (Federation Cynologique Internationale 2017a) are used to ensure enough space for the dogs to perform individual preferred inter-hurdle strides, set-off and landing angle without restrictions (Birch *et al.* 2015; Birch & Lesniak 2013). A pole was placed 7 meters in front of the first jump and 7 meters after the last jump to mark where the dog should start and end to ensure enough space to set-off and land and to get a clean straight run without turning around while jumping. This is done for two main reasons. First concerning security, it was done to ensure the dogs did not jump close to the wings of the hurdles with the risk of getting the AMG-equipment entangled, and second to ensure that the measurements of the muscles did not get affected by turns or deceleration. The bars were positioned at 3 heights, 45 cm as the lowest, 55 cm as the



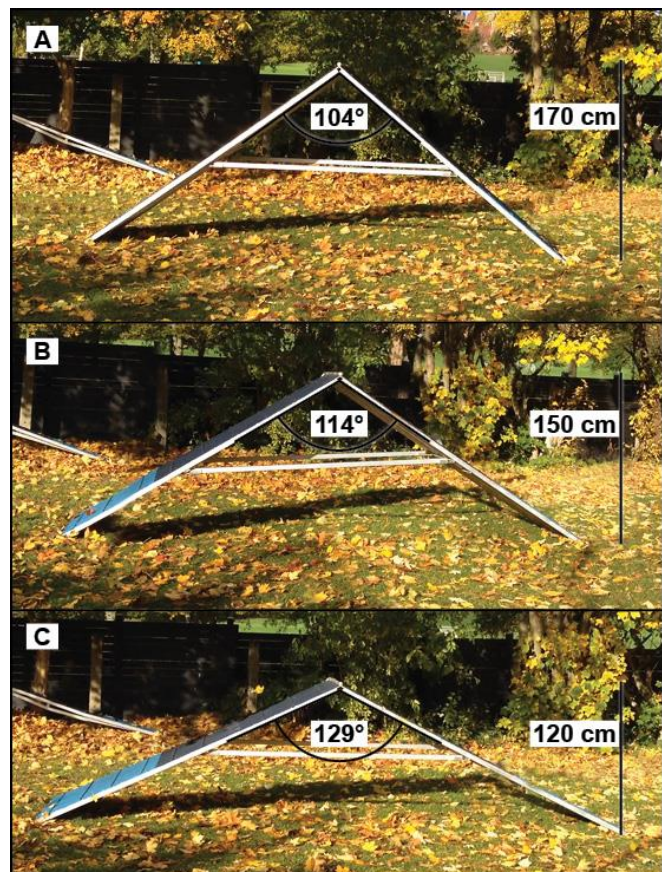
medium and 65 cm as the highest. Although it would be easier to have holders in each height, only one holder was positioned on each wing to avoid risk of injuries and entanglement of the AMG-equipment in the exposed holders.



Picture 14: The position of the exercise equipment on the training area belonging to The Danish Agility Association Copenhagen at the address Lersøstien 175, 2100 Copenhagen East.

### 2.5.3 The A-frame

The A-frame used in this experiment had 3 possible heights besides lying flat on the ground. The heights were 170 cm, 150 cm and 120 cm with an angle under apex of  $104^\circ$ ,  $114^\circ$ ,  $129^\circ$  respectively as illustrated in picture 15. The ramp going up and downhill were each 2.72 meters resulting in a total length of the run over the A-frame of 5.44 meters no matter the height. When positioned in the highest position a pole was placed at a distance of 7 meters from each side of the A-frame to locate where to start and stop the run. This was done to ensure an even run without the dog turning to go back up the A-frame immediately or turning to look for the owner and hence affect the measurements of the muscles.



Picture 15: The three heights of the A-frame and associated angles under apex for the position of A: high A-frame, B: medium A-frame and C: low A-frame.

## 2.5.4 The AMG-equipment

AMG-equipment were used to measure how the muscles of the dog is affected by the different exercises.

The CURO was placed in the handle of a Julius-K9<sup>®</sup> harness for dogs. To ensure the harness fit the different sizes of large dogs the sizes 0, 1 and 2 was available. Two sensors were used per dog as well as EKO GEL and 4 pieces of Snøgg of approximately 15 x 15 cm.

An Ipad 2 was bought and the program CURO Equine with the update from June 6<sup>th</sup> 2016 was installed.

## 2.6 Method

### 2.6.1 Literature search

Literature was found using the databases Web of Science, Ovid<sup>®</sup> (Medline and CAB Abstract), Google Scholar and the Royal Danish Library – Copenhagen. In the database for scientific articles, the search words and combinations were: *>dog\* or canine\* or pup\* <, > horse\* or equine\* <, >jump\* OR hurdle\* OR agility\* OR A-frame\* OR obstacle\* OR sport\* <, >warm\* AND up\* OR warm-up\*, cool\* down\* OR post\* exercise\* OR exercise\* OR cool\* OR post-exercise\* OR cool-down\* OR post work out\* OR recovery\* exercise\* <, >lactic acid\* or lactose\* or pyruvat\* <, >muscle\* OR exercise\* OR recov\* <, > training\* OR exercise\* OR training level\* OR fitness\* <, >acoustic myography OR bioimpedance OR electromyography <, >neuro\* or nerve\* or muscular\* or impulse\* <, >muscle\* OR muscular\* OR fib\* OR fib\* type\* <, >elastic\* AND energy\* OR elastic strain energy <, >eccentric\* OR concentric\* <, > trot\* or gallop\* or walk\* <, > triceps brachii OR triceps\* <, >gluteus superficialis OR gluteus\* <, >biomechanic\* OR move\* OR motion\* <, >Injur\*OR damage\* OR fracture\* OR trauma\* <*. Some were shortened, but all were combined with “OR” or “and” in different combinations using multi-field search. From articles matching the search criteria similar articles, citing articles and articles cited were used. If cited articles could not be found in the databases for scientific articles Google Scholar were used or the articles were ordered home from the Royal Danish Library - Copenhagen as well as references to books. Non-scientific knowledge such as regulations were found using Google. Textbook material was used for basic knowledge of muscle and nerve composition and function.

### 2.6.2 Before the start

It is recommended to look at appendix F for a further understanding of the procedure of the exercises and what was noted for each exercise. All exercises were video recorded on an I-Phone 4 or 5 video camera except for the walk in the park.



Before the handler and the dog arrived, leaves were swept away and the agility equipment was placed as described previously. According to weather it was decided that the experiment would be completed if there was only a little rain and the ground was not too slippery. The weather and condition of the ground was noted in the protocol. Before the exercises began the handler signed a contract with acceptance of the data being used for the thesis and acceptance of the author disclaiming responsibilities for injuries (appendix G).

### 2.6.3 The mounting of AMG-equipment

The mounting of the AMG-equipment was done with as little discomfort as possible for the dog. To ease the mounting of the AMG-equipment the dog were told to jump unto a table unless this was associated with discomfort by the dog. This was done to avoid leaning over the dog making the dog uncomfortable and to make the working position more comfortable for the humans.

The owner was offered the 3 sizes of harnesses and told to adjust the harness to the dog's size. This was done to make the positioning and introduction of the harness less threatening compared to an unknown person coming with a new harness. The positioning of the harness was checked according to the recommendation by Julius-K9® (Julius-K9® 2016).



Picture 16: The position of the CURO (MyoDynamik ApS, Frederiksberg, Denmark) under the handle of a Julius-K9 harness (Julius-K9®, Szigetszentmiklós, Hungary)



Picture 17: The position of the sensor on gluteus superficialis between the iliac crest (position of thumb) and the ischial tuberosity (position of middle finger).



Picture 18: The position of the sensors on a 15 x 15 cm piece of Snøgg after use explaining the fur.



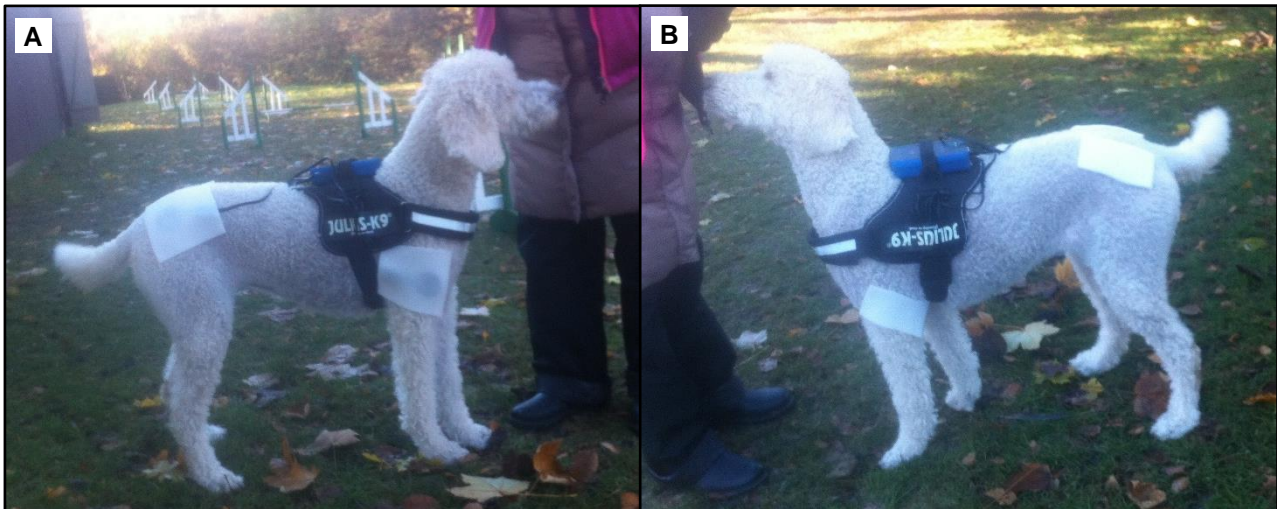
Picture 19: The position of the sensor on triceps brachii between the proximal humerus and the olecranon of the ulna.

The CURO was positioned under the handle of the harness (picture 16), and the sensors were placed on the right side of the dog as follows. First the sensor at gluteus superficialis was mounted as the handling of the dog from behind is less threatening than from up front. Gluteus superficialis was found between the iliac crest and the ischial tuberosity and could be palpated, picture 17. The fur was divided and gel was massaged onto the visible skin over an area large enough to cover the plates of the sensors. The ultra-sound gel EKO GEL was applied to the sensors both front and back. The sensor was positioned on a piece of Snøgg with the receiving plate visible (picture 18). The sensor with the Snøgg was placed on the muscle and the Snøgg was fastened to the fur. The connecting wire was inserted into the R1 gate on the CURO, and the wire was folded and positioned under the Velcro on the side of the harness without interfering with the mobility of the dog. The same procedure was done when positioning the sensor at triceps brachii. Triceps brachii was found between the proximal humerus and the olecranon of the ulna and the sensor was placed closest to the olecranon of the ulna due to the need of distance from the harness (picture 19). The sensor was connected to the L1 gate on the CURO.

Additional pieces of Snøgg were positioned on the same location on the left side of the dog (picture 20) to ensure that the dog did not walk unevenly or relieve the muscles where the sensors were positioned, which could affect the measurements.

The CURO was turned on and using Wi-Fi, connected to the Ipad with the CURO Equine App. The dog was positioned in a static position and the harness pulled to the sides and forth and back to ensure no noise was detected by the sensors due to the harness. The dog walked a few meters to secure correct measurements shown on the Ipad. All devices using Wi-Fi were turned off or set on flight-mode to avoid interference with the data sent from the CURO to the Ipad.

After the mounting of the AMG-equipment, the dog could start the warm-up exercises.



Picture 20: The position of the CURO in the handle on the Julius-K9® harness and adjacent equipment. A: Right side of the dog with sensors positioned under the Snøgg and the wires fastened under the Velcro on the Julius-K9® harness. B: Left side of the dog with the position of Snøgg equivalent to the right side, but without the sensors.

#### 2.6.4 The warm-up

To investigate how the muscles are affected by warm-up and the amount of time used for warm-up, a 5 minute long trial of active general and specific warm-up exercises was repeated 3 times. Before the first trial and after each trial a short trot was performed for the purpose of comparison of the muscles. Each trial comprised of; 1 minute brisk walking; 1 minute trotting, 0.5 minute gallop; 1 minute practice of shifting side according to the handler while walking in a large circle; 0.5 minute slalom between legs while the owner walked; 0.5 minute figure-eight movements between the owners legs while the owner stood still; and 0.5 minute for 3 times walking the cavaletti, in total 5 minutes. The time allocated were guidelines and could be shortened or lengthened with a few seconds according to the efficiency of the handler and dog performing the exercise. If the dog jumped all the way through the cavaletti it did not count as a correct exercise and the dog had a new attempt. Between each of the three trials the dog had a 2-minute break and was offered water.

The CURO was turned on before each trial and turned off after each trial. This was done to ensure no large files or long recordings as well as making the division of the exercises easier when looking



at the graphs on the computer. The time the dog started each exercise was noted and video recordings were made on an iPhone-camera when the CURO was recording. There was a total of 4 recordings.

After the warm-up, the handler was instructed in how to do the next exercise.

### 2.6.5 The hurdles

To investigate how the muscles are affected by jump height the dog was presented with five hurdles at 3 different heights as described in the section about materials. A trial comprised of a dog jumping 5 hurdles at equal height. This was done for the three heights from the lowest to the highest position. This sequence was repeated two more times, resulting in 9 trials. The procedure can be seen in the protocol appendix F. The same procedure and order was followed for all dogs.

The handler could freely choose what method they would like to use when sending their dog forward, e.g. run beside the dog and command the dog forward, position the dog in front of the first hurdle and stand at the other end of the 5 hurdles calling the dog, or holding the dog in front of the first hurdle and having a person standing at the other end calling the dog.

For the same reasons, as with the warm-up the CURO was turned on before each trial of 5 jumps and turned off after each trial, giving a total of 9 recordings.

This sequence of the trials according to heights was chosen to avoid muscle fatigue affecting the results for all the trials in one height. There may be muscle fatigue in the later trials, but it is hypothesized that this will be seen on the ESTi<sup>TM</sup>-scores and taken into account. It is expected that muscles do not fatigue after the 3 first trials and according to the shape of the dog it should not fatigue significantly over the 9 trials.

After the 9 trials of jumping, the dog took a rest while the handler was instructed in how to perform the next exercise.

### 2.6.6 The A-frame

Before performing the exercise, the handler had decided what kind of contact method they would practice. A trial comprised of a dog running the A-frame three times, starting at the pole in front of the A-frame and ending at the pole after the A-frame between each run. This was done for 4 trials. First with the A-frame in high position, next medium position, thirdly low position and finally at high position practicing other contact method. If they had practiced the 2o2o-contact method in the first 3 trails, they were told to practice running contacts in the last trial, and opposite. The chosen contact method was only performed when running down the A-frame. The 3 runs in the 4 trials resulted in a total of 12 runs. The same procedure and order was followed for all dogs. If the

dog had difficulties when practicing the two-on-two-off method aid was offered which could be in the form of a bowl with a treat strategically positioned approximately 30-50 cm after the A-frame according to the size of the dog.

The CURO was turned on before each trial consisting of 3 runs of the A-frame and turned off after each trial, giving a total of 4 recordings.

This sequence of the trials according to height, was chosen due to the labour requiring work of adjusting the height of the A-frame.

### 2.6.7 The cool-down

To investigate how the muscles are affected by cool-down, the dog performed a series of exercises as follows: 2 minutes of casual trot, 1 minute of figure-eight movements between the owner's legs while the owner stood still, followed by a brisk walk for 2 minutes. After this the dog was put on a leash and taken for a regular walk in the park for 10 minutes. A total of 15 minutes for cool-down. After the walk in the park the dog ran a short interval of trot, so as to compare the trot at the beginning with the trot after the cool-down.

The CURO was turned on before the first exercise and turned off before going to the park, and turned on and off again in connection with recording of the last trot, resulting in two recordings.

### 2.6.8 The removal of AMG-equipment

The removal of the AMG-equipment was done with as little discomfort as possible for the dog. Again, the dog was placed on a table, unless this was associated with discomfort by the dog. After turning the Wi-Fi and CURO off, the Snøgg was gently peeled off the fur together with the sensors, and the sensors were detached from the CURO and cleaned. The harness was loosened and together with the CURO removed from the dog. When the AMG-equipment was removed, the dog was allowed to move freely.

### 2.6.9 The questionnaire

Each participant was asked a set of questions about their dog for the purpose of comparing individual results. See the questionnaire in appendix H. The questions addressed three main areas: regular information, health status and agility training. First regular information was assessed: name, gender, status eg. castrated/sterilized, breed, date of birth, height over withers and weight. When the owner of the dogs was uncertain about the height it was measured on location. The body condition was scored by the author with the traditional score from 1 to 9, 1 being very thin and 9 very fat (appendix I). The sportiness of the dog was assessed using a score from 1 to 5 with 1 being a sporty dog illustrated with a border collie and 5 being a large and heavy dog illustrated

with a Saint Bernard. Second the health status was assessed by questioning the current and earlier status of injuries, and if so when and where the injury was located and the cause. No injured dog was allowed to participate in the experiment. Knowledge about the use of alternative preventive and treatment methods such as: physiotherapy, massage, chiropractor, acupuncture was assessed. Thirdly and last it was asked how long the dog had practiced agility and when they last had trained agility. The level of training the last three months, whether or not they use warm-up and cool-down exercises in connection with training and competitions and for how long, was assessed. The normal height of the hurdles when training and the training method on contact with the A-frame was noted.

This information was gathered for each dog in connection with the execution of the experiment. In total the briefing of the owners, the mounting of the CURO, executing the exercises, the questionnaire, the removal of the CURO and the debriefing took approximately 1.5 to 2 hours, and less time if an owner came with two or more dogs. To ensure no disturbance from new dogs coming to the area, and a buffer for complications, 2 to 2.5 hours was assigned to each dog.

## 2.7 Data processing

After each day with trials the raw data were transferred from the CURO to the computer from where it was later uploaded to the homepage <http://curo.softtheme.com/site/login> to be analysed. When a score of 0 was achieved on all the ESTi™-parameters the markings were changed without compromising the selection criteria. When still no score above 0 was achieved the data were omitted. For each dog the parameters E, S and T for both muscles were found and listed in Excel according to exercise and the ESTi-scores were calculated. For those exercises where more than one measurement was conducted (hurdles and A-frame) an average of the ESTi™-scores was calculated to simplify the data for further tests. For all data sets in the following description an average, median, minimum, maximum and standard deviation of all the dogs were calculated. Radar diagrams were used to present the data as it is more visual and easier to interpret.

When the data could not be analysed, for example all parameters were zero, they were excluded from the results. Ninety-three of 3276 results were excluded. Some sections of soundwaves for analyses had a higher amplitude than the sensors could measure which resulted in the top of the amplitude being clipped and the S-score being zero. As illustrated on picture 22 this could be seen on triceps on dogs running up the A-frame.

### 2.7.1 The warm-up exercise

A minimum of 5 seconds of a regular trotting session was located in the recording before the first warm-up exercise and correspondingly in the end of each of the three recordings of warm-up exercises. The parameters for analysis of trot was setup as shown in table 7. There were noted 24 numbers per dog for further analysis.

Table 7: Setup of AMG parameters for analysis of trot and jumping.

Parameters	Setup values
Threshold (mV)	0.20
Max S (mV)	0.99
Max T (Hz)	120
CST (sec)	0.15

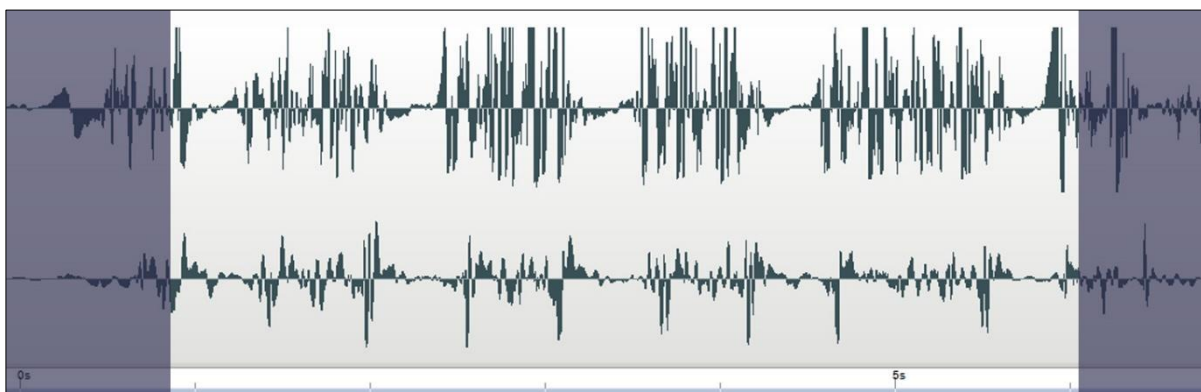
Eight boxplots were made comparing before warm-up exercises, 5 minutes of exercises, 10 minutes of exercises and 15 minutes of exercises respectively for the four sets of ESTi™-scores for each muscle. The boxplots are presented in section 3.1 *The warm-up* and the data used can be found in Appendix J.

### 2.7.2 The hurdles

For each recording consisting of 5 jumps a section was analysed from the set-off at the first hurdle until after the impact of landing after the fifth hurdle as shown in picture 21. The parameters for analysis of jumping was setup as shown in table 7. There were noted 54 numbers per dog for further analysis.

To simplify the data for further tests the average of the ESTi™-scores in the three heights for each dog was calculated and the data used can be found in Appendix K.

Boxplots of the recorded ESTi™-scores on triceps brachii and gluteus superficialis respectively when jumping the three heights was made. The boxplots are presented in section 3.2.1 *Triceps brachii* and 3.2.2 *Gluteus superficialis*.



Picture 21: Section of five jumps for analysis marked.

### 2.7.3 The A-frame

Each recording consisted of the dog running the A-frame three times, and each run of the A-frame was analysed separately. For each run of the A-frame the run up and down was analysed separately. The session of the run up the A-frame was analysed from the measured leg had the first contact with the A-frame until and included the last contact with the A-frame going uphill. When going over the apex the

muscle activity was visibly lower as no legs touched the A-frame. The run down the A-frame was analysed from the point where the leg for the measured muscle had the first contact with the A-frame downhill until the dog stood still or had contact with the ground, depending on the chosen contact method. This is illustrated in picture 22 and 23. The parameters for analysis of the muscle activity on the A-frame was setup as shown in table 8. A standard setup was initially chosen, but the setup for threshold was not suitable for all dogs, muscles nor both running up and down.

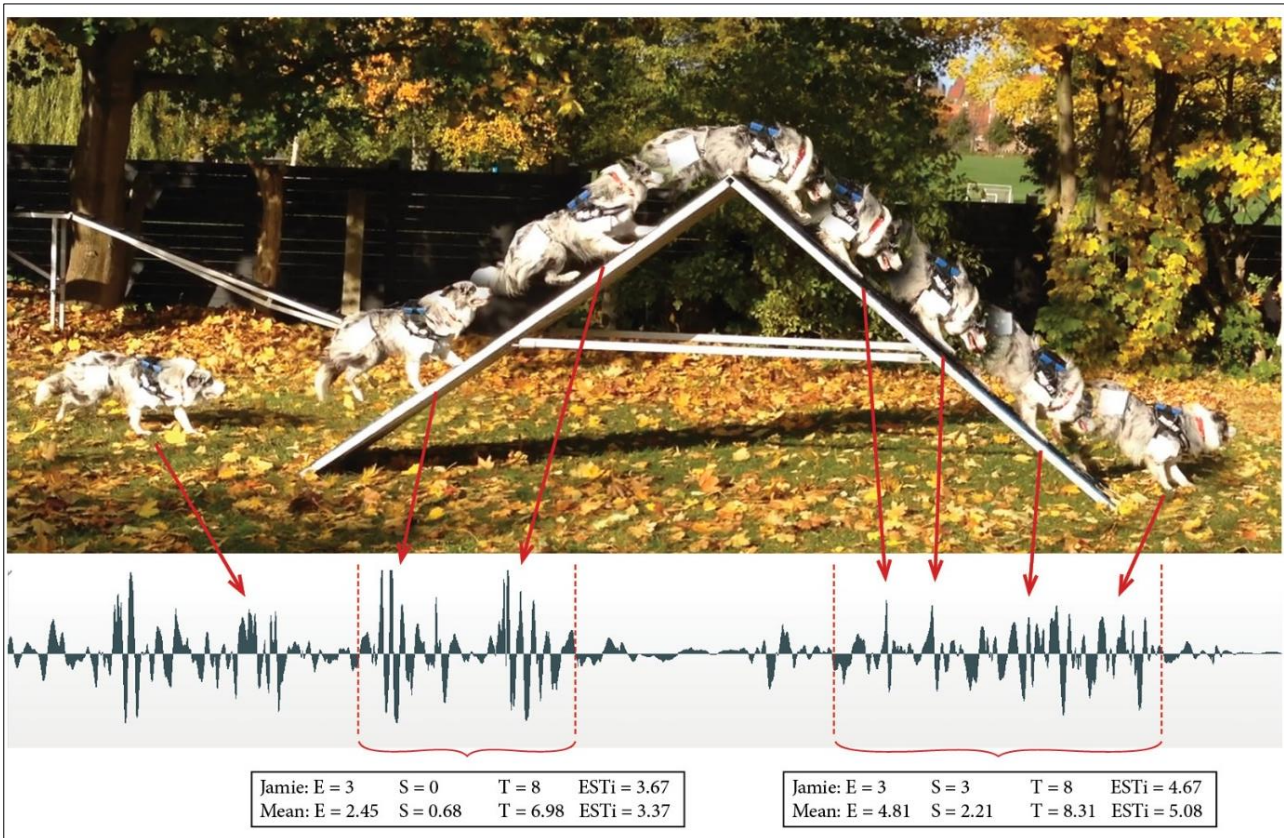
When the initial setup was not sufficient to catch the soundwaves the threshold setup was manually changed until an ESTi-score was obtained. There were noted 144 numbers per dog for further analysis.

To simplify the data for further tests the average of the ESTi<sup>TM</sup>-scores in the three heights and when changing contacts method was calculated for each dog as presented in appendix L.

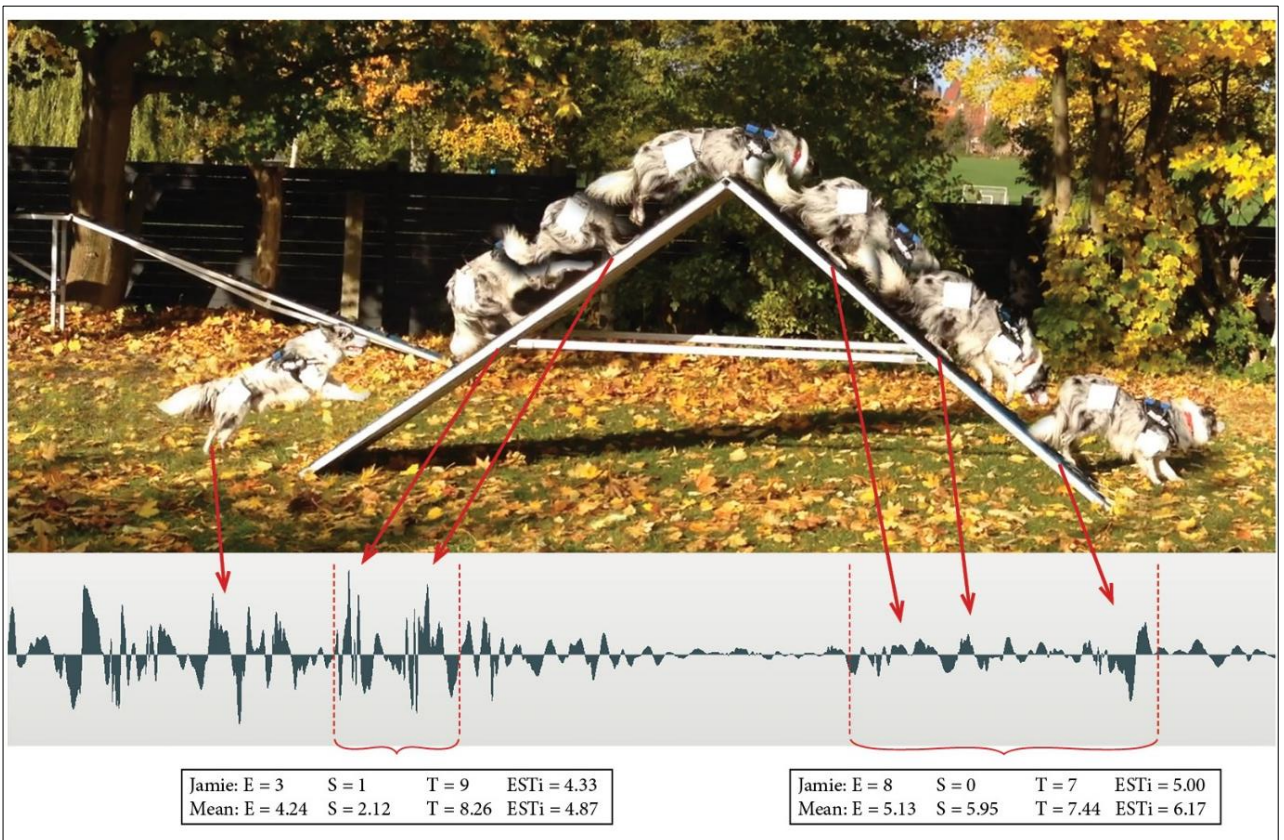
Table 8: Setup of AMG parameters for analysis of gluteus superficialis (GS) and triceps (TC) when running up and down the A-frame.

Parameters	Setup values
Threshold (mV)	Standard (alternative)
TC down and GS up	0.50, (0.40)
TC up	0.70, (0.50)
GS down	0.55, (0.20, 0.10, 0.05)
Max S (mV)	0.99
Max T (Hz)	120
CST (sec)	0.15





Picture 22: Measurements of triceps with the use of acoustic myography on a dog (Jamie) running the A-frame positioned high with 2on-2off contact method.



Picture 23: Measurements of gluteus superficialis with the use of acoustic myography on a dog (Jamie) running the A-frame positioned high with 2on-2off contact method.

#### 2.7.4 The cool-down exercise

A minimum of 5 seconds of a regular trotting session was located in the recording before the cool-down exercises and correspondingly in the end of the recording of the cool-down exercises. The parameters for analysis of trot was similar with the warm-up as shown in table 7. There were noted 12 numbers per dog for further analysis.

## 2.8 Statistics

Real Statistic Resource Pack for Excel 2010/2013/2016 for Windows (version 4.13, 2010, Real Statistics Using Excel, Charles Zaiontz) was downloaded from the homepage <http://www.real-statistics.com/free-download/> and installed in Excel as an add-in pack. This made it possible to make more advanced statistic tests and analysis of data which are normally not available in Excel.

The ESTi<sup>TM</sup>-scores were compared in pairs to test statistical difference between the averages for the hurdles, the A-frame and the cool-down exercises. Firstly, a Shapiro-Wilk test was conducted to ensure the data were normally distributed. For the normally distributed data a two-tailed paired sample t-test was conducted. For data, not being normally distributed, a Wilcoxon signed ranks test for paired samples with two tails was conducted. ANOVA was used for the warm-up exercises and the Wilcoxon Rank-Sum test was used for the descriptive data as explained in the following. P-values below 0.05 were considered significant.

#### 2.8.1 The warm-up exercise

Equality between the average ESTi<sup>TM</sup>-scores before warm-up exercises, 5 minutes of exercises, 10 minutes of exercises and 15 minutes of exercises, were tested using Repeated Measures ANOVA for the data being normally distributed. The data not being normally distributed was tested using One-way ANOVA. This was done in the program GraphPadInStat 3 for Mac (Version 3.0b, 2003, GraphPad Software Inc., La Jolla, California). This resulted in 80 tests. The result of the tests can be seen in appendix J.

#### 2.8.2 The hurdles

Equality between the average ESTi<sup>TM</sup>-scores for low, medium and high hurdles were tested pairwise against each other. Due to the risk of confounding the effect from fatigue or improvement the equality between the average ESTi<sup>TM</sup>-scores from first to last run was tested for all heights. This resulted in 120 tests. The result of the tests can be seen in appendix K.



### 2.8.3 The A-frame

Equality between the average ESTi™-scores for low, medium and high A-frame were tested pairwise against each other. Likewise, the two practice of contact were tested against each other. The same tests were made when clustering the 11 dogs practicing the 2o2o-contact method together and the 3 dogs practicing the running contact method together. Due to the risk of confounding the effect from fatigue or improvement the equality between the average ESTi™-scores from first to last run was tested for all heights. This resulted in 832 tests. The result of the tests can be seen in appendix L.

### 2.8.4 The cool-down exercise

Equality between the average ESTi™-scores before and after cool-down exercises were tested pairwise against each other. This resulted in 24 tests. The result of the tests can be seen in appendix M.

### 2.8.5 The descriptive data

To test whether or not the results were affected by different characteristics of the dogs, the dogs were clustered in two groups as shown in table 9. Body condition score was not considered as the dogs were similar with only one score of difference from highest to lowest score. The status of dogs being intact was neither considered, as the clusters would be too small for each gender and it were not expected to have a significant effect on the results. The dogs were divided for each character as listed in table 9. The division was partly based on the median (see table 5 and 6) to avoid small populations, and partly on normal sense e.g. gender. The use of athletic score was incorporated instead of breed, because the variation of physical appearance within a breed may be significant. As an example, both a slim and broad Australian shepherd were represented.

Table 9: Clustering of the 14 experimental dogs according to gender, if the dog is intact or castrated/neutralized, height to the withers, weight, age, score of how athletic the dog is built on a scale from 1-5, years of experience in agility, the training level measured in how many hours a week the dog practice, the height of the hurdle in normal training situation and the practice of contact method when running the A-frame being either the 2on-2off contact method (2o2o) or running contact method (running).

Character	Cluster 1 (n)	Cluster 2 (n)
Gender	Male (6)	Bitch (8)
Height	≤ 55 cm (7)	> 55 cm (7)
Weight	≤ 21 kg (7)	> 21 kg (7)
Age	≤ 3 years (7)	> 3 years (7)
Athletic score	= 1 (5)	≥ 1 (9)
Experience	< 2 years (5)	≥ 2 years (9)
Training level	< 1 hour a week (8)	≥ 1-2 hours a week (6)
Normal hurdle height	< 55 cm (4)	≥ 55 cm (10)
Normal contact method	2o2o (11)	Running (3)

Due to the extensive data work and the small population size these tests were not tested for normality. All tests conducted were Wilcoxon Rank-Sum tests, as the data are not paired and considered not to be normally distributed. The test of difference between the clusters was made for all the experiments (the warm-up exercise, the hurdle height, the height of the A-frame and the cool-down exercise). This resulted in 1224 tests. The result of the tests can be seen in appendix J, K, L and M.

## 3.0 Results

### 3.1 The warm-up exercise

#### 3.1.1 Triceps brachii

Figure 1 shows a significant difference in the E-score between no time spent on warm-up exercises compared to 5, 10 and 15 minutes spent on warm-up exercises respectively. The more time the dog spent on warm-up exercises the more significant the differences were. No significant difference was found between 5, 10 and 15 minutes of warm-up exercises for the E-score. Overall the dogs were better at using triceps brachii after 10 and 15 minutes of warm-up exercises compared to before doing warm-up exercises as the ESTi-score is significantly increased. This is also illustrated in figure 2 with a radar diagram which is more visual and easier to interpret. An overall combined score can be seen, as well as an optimal scale of where the dogs are.

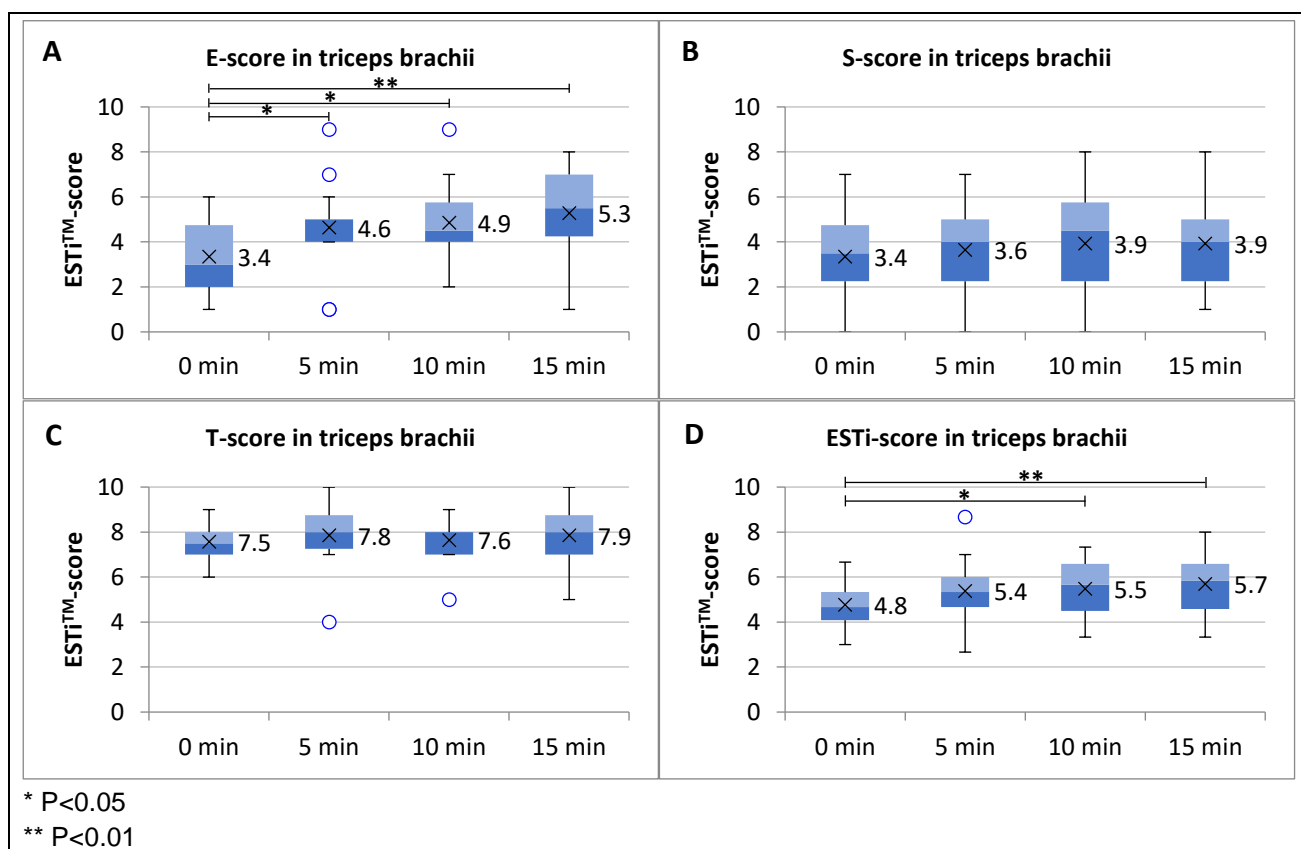


Figure 1: Boxplot of the recorded ESTi™-scores on triceps brachii (n = 14) after 0, 5, 10 and 15 minutes of warm-up exercises. The X-axis shows the time spent on warm-up exercises and the y-axis show the average ESTi™-score. The X's and adjacent number mark the mean, while the border between the light blue and the blue box marks the median. The top of the light blue box marks the 3rd quartile while the bottom of the blue box marks the 1st quartile. The top and bottom of the whiskers marks the maximum and minimum of the data set respectively. The blue circles marks outliers. Panel A shows the E-score in triceps brachii and a Repeated Measures ANOVA was conducted to test for significant difference. Panel B shows the S-score in triceps brachii and a repeated measures ANOVA was conducted to test for significant difference. Panel C shows the T-score in triceps brachii and a one-way analysis of variance was conducted to test for significant difference due to data not being normal distributed. Panel D shows the ESTi-score in triceps brachii and a repeated measures ANOVA was conducted to test for significant difference. Data used for the boxplots for triceps brachii can be found in Appendix J.

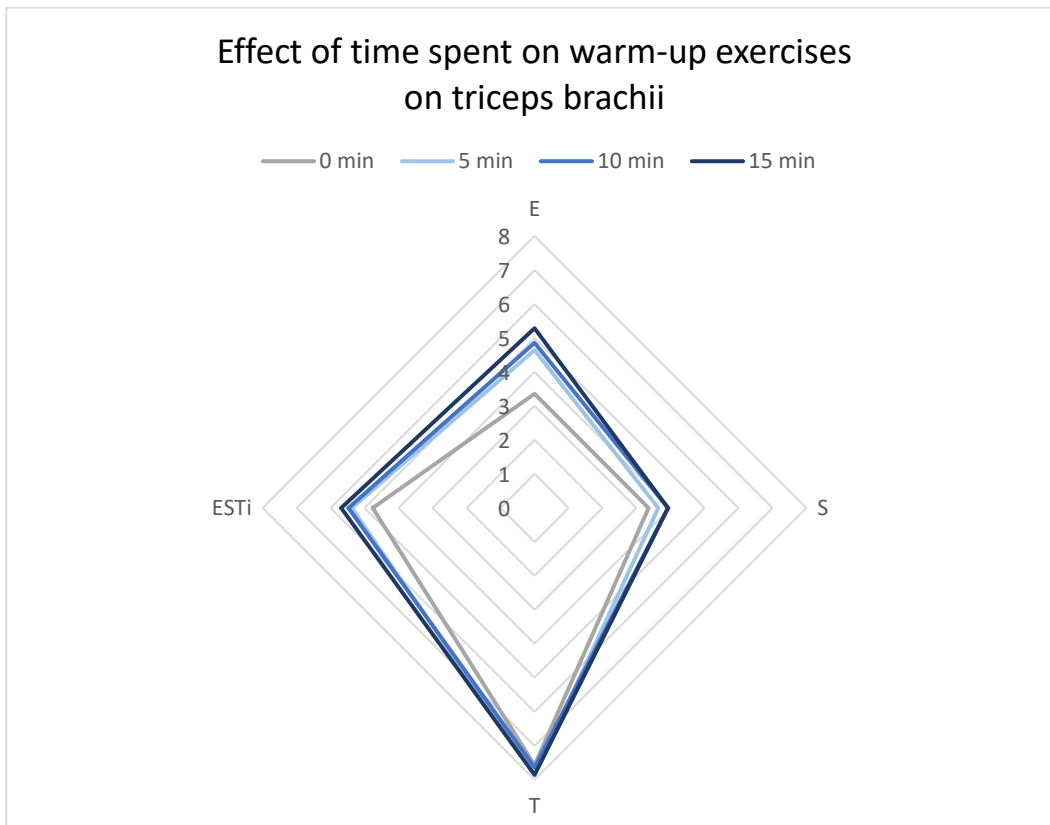


Figure 2: Radar diagram of the average recorded ESTi™-scores on triceps brachii (n = 14) after 0 (grey line), 5 (light blue line), 10 (blue line) and 15 (dark blue line) minutes of warm-up exercises. The axis from center and out shows the ESTi™-score. Data used for the radar diagram for triceps brachii can be found in Appendix J.

### 3.1.2 Gluteus superficialis

There were in general no significant difference between the time spent on warm-up exercises on gluteus superficialis according to the ESTi<sup>TM</sup>-scores, see figure 3. Figure 4 confirm this more visually.

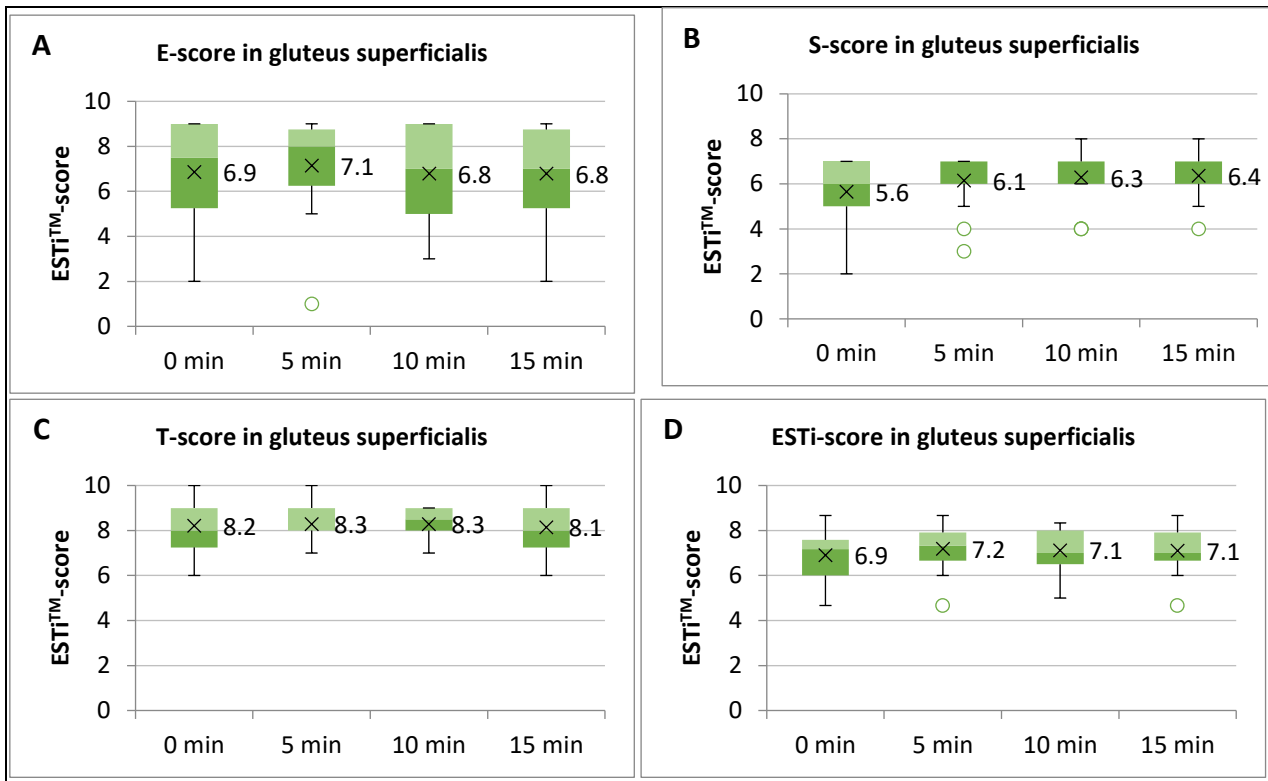


Figure 3: Boxplot of the recorded ESTi<sup>TM</sup>-scores on gluteus superficialis ( $n = 14$ ) after 0, 5, 10 and 15 minutes of warm-up exercises. The X-axis shows the time spent on warm-up exercises and the y-axis show the average ESTi<sup>TM</sup>-score. The X's and adjacent number mark the mean, while the border between the light green and the green box marks the median. The top of the light green box marks the 3rd quartile while the bottom of the green box marks the 1st quartile. The top and bottom of the whiskers marks the maximum and minimum of the data set respectively. The green circles marks outliers. Panel A shows the E-score in gluteus superficialis and a one-way ANOVA was conducted to test for significant difference due to data not being normal distributed. Panel B shows the S-score in gluteus superficialis and a one-way ANOVA was conducted to test for significant difference. Panel C shows the T-score in gluteus superficialis and a one-way analysis of variance was conducted to test for significant difference. Panel D shows the ESTi-score in gluteus superficialis and a repeated measures ANOVA was conducted to test for significant difference. Data used for the boxplots for gluteus superficialis can be found in Appendix J.

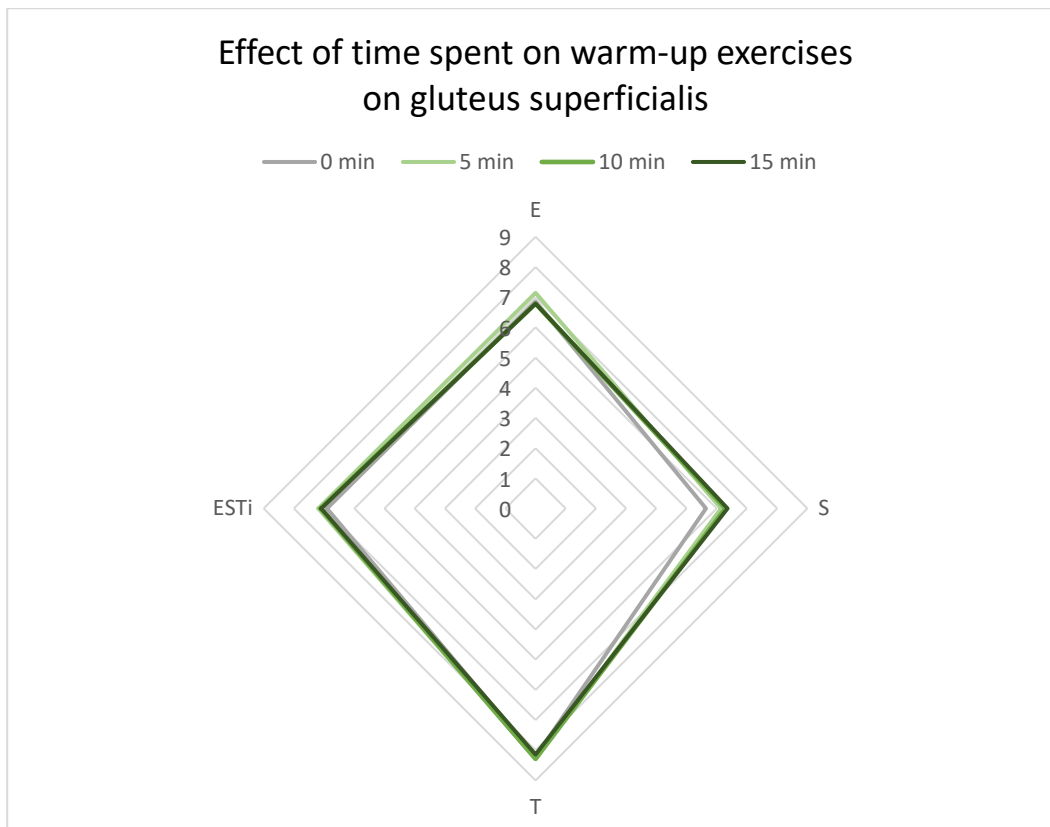


Figure 4: Radar diagram of the average recorded ESTi™-scores on gluteus superficialis (n = 14) after 0 (grey line), 5 (light green line), 10 (green line) and 15 (dark green line) minutes of warm-up exercises. The axis from center and out shows the ESTi™-score. Data used for the radar diagram for gluteus superficialis can be found in Appendix J.

### 3.1.3 The descriptive data

No significant difference was found when clustering the dogs according to gender, weight, age, normal hurdle height in training and normal contact-method practiced on A-frame. Only few significant differences with no obvious connection was found when clustering according to height, experience or athletic score.

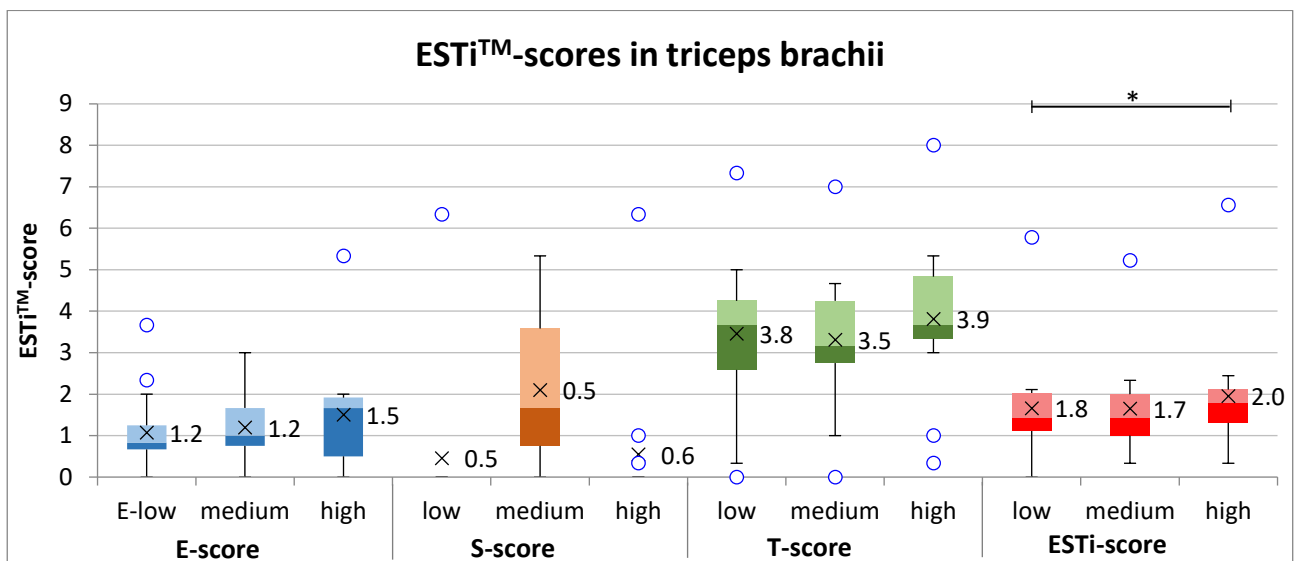
According to training level the dogs with 1 hour of training per week or more (n=6) had a significantly higher ( $p < 0.05$ ) S-score for triceps brachii after both 10 (S=5.7) and 15 (S=5.3) minutes of warm-up exercises compared to dogs with less than 1 hour of training per week (n=8) (10 and 15 minutes S=2.6, S=2.8 respectively). There was no significant difference before the warm-up exercises or after 5 minutes of warm-up exercises. Dogs with 1 hour of training per week or more recruited less muscle fibers in triceps brachii after 10 and 15 minutes of warm-up exercises compared to dogs with less than 1 hour of training per week. No significant difference was found in gluteus superficialis. See appendix J for data.



## 3.2 The Hurdles

### 3.2.1 Triceps brachii

A significant difference between the different heights of the hurdles were found when looking at triceps brachii according to the ESTi™-scores, see figure 5. There was a significant increase in the ESTi-score in triceps brachii when jumping high compared to low hurdles with an increase of the ESTi-score from 1.8 to 2.0. This means that in an overall assessment the dogs, triceps brachii was more coordinated and effective, when jumping high hurdles compared to low hurdles. This can also be seen in figure 6. No significant difference was found when testing the first run with the last run for each height, meaning no sign of fatigue was found.



\* P<0.05

Figure 5: Boxplot of the recorded ESTi™-scores on triceps brachii on dogs (n = 14) when jumping hurdles with the heights of 45 cm (low), 55 cm (medium) and 65 cm (high) respectively. The X-axis shows which height is observed for each ESTi™-score and the y-axis show the average ESTi™-score. The X's and adjacent number mark the mean, while the border between the light-colored and the darker-colored boxes marks the median. The top of the light-colored box marks the 3rd quartile while the bottom of the darker-colored box marks the 1st quartile. The top and bottom of the whiskers marks the maximum and minimum of the data set respectively. The blue circles marks outliers. A paired two-tailed t-test was conducted to find significant difference for the T-score, but for the comparison of data for the E, S and ESTi-score a Wilcoxon Signed-Ranks Test was conducted due to data not being normally distributed. Data used for the boxplots for triceps brachii can be found in Appendix L.

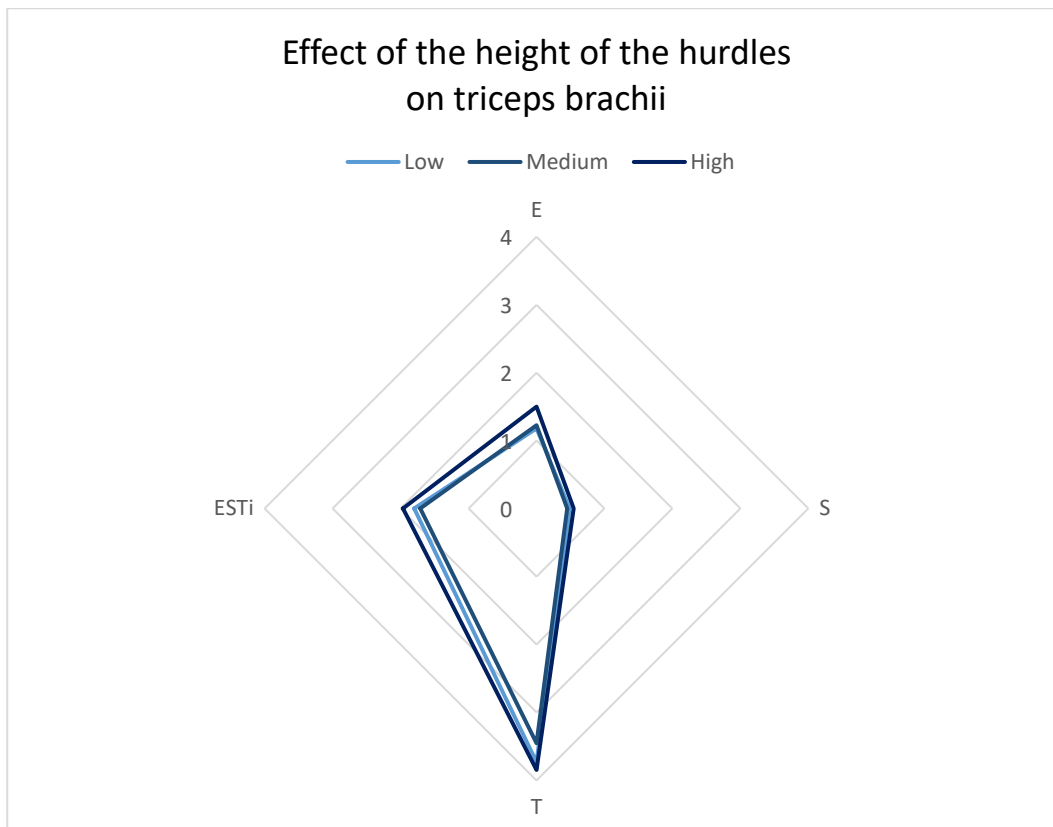
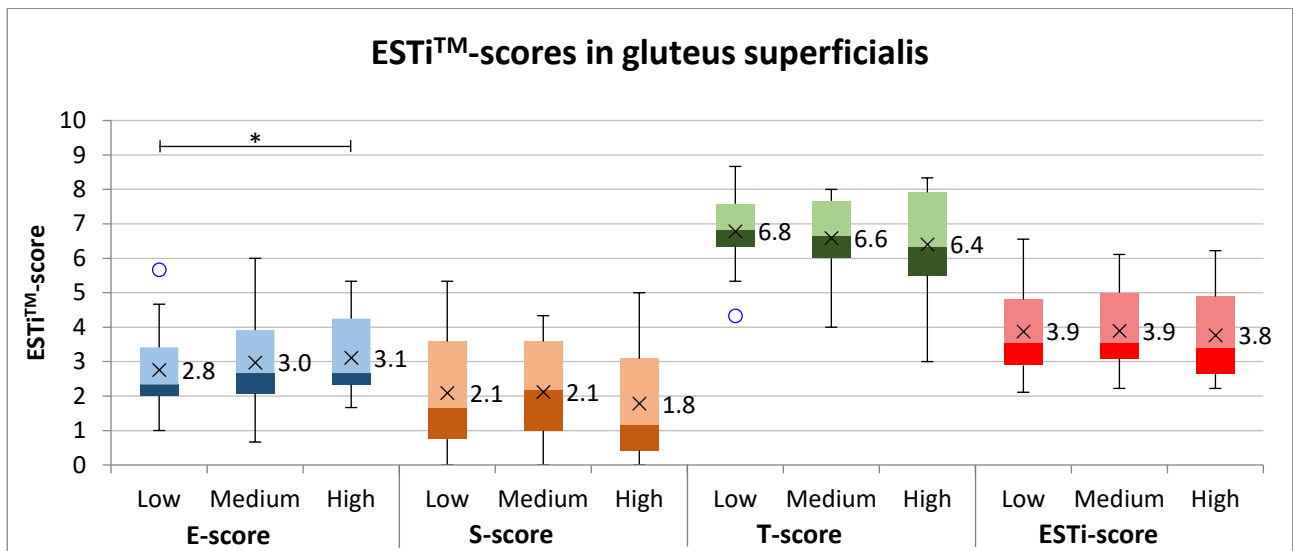


Figure 6: Radar diagram of the average recorded ESTi™-scores on triceps brachii on dogs (n = 14) when jumping hurdles with the heights of 45 cm (low = light blue line), 55 cm (medium = blue line) and 65 cm (high = dark blue line) respectively. The axis from center and out shows the ESTi™-score. Data used for the radar diagram for triceps brachii can be found in Appendix L.

### 3.2.2 Gluteus superficialis

Some significant differences between the different heights of the hurdles were found when looking at gluteus superficialis according to the ESTi™-scores, see figure 7. There was a significant increase in the E-score in gluteus superficialis when jumping high compared to low hurdles with an increase of the E-score from 2.8 to 3.1. This means that the dogs used gluteus superficialis more efficient when jumping high hurdles compared to low hurdles. Figure 8 visualizes the results. No significant difference was found when testing the first run with the last run for each height, meaning no sign of fatigue was found.



\* P<0.05

Figure 7: Boxplot of the recorded ESTi™-scores on gluteus superficialis on dogs (n = 14) when jumping hurdles with the heights of 45 cm (low), 55 cm (medium) and 65 cm (high) respectively. The X-axis shows which height is observed for each ESTi™-score and the y-axis show the average ESTi™-score. The X's and adjacent number mark the mean, while the border between the light-colored and the darker-colored boxes marks the median. The top of the light-colored box marks the 3rd quartile while the bottom of the darker-colored box marks the 1st quartile. The top and bottom of the whiskers marks the maximum and minimum of the data set respectively. The blue circles marks outliers. A paired two-tailed t-test was conducted to find significant difference for the E and S-score except the comparison of low to medium for the S-score. The low to medium for the S-score as well as all T- and ESTi-scores were tested with a Wilcoxon Signed-Ranks Test due to data not being normally distributed. Data used for the boxplots for gluteus superficialis can be found in Appendix L.

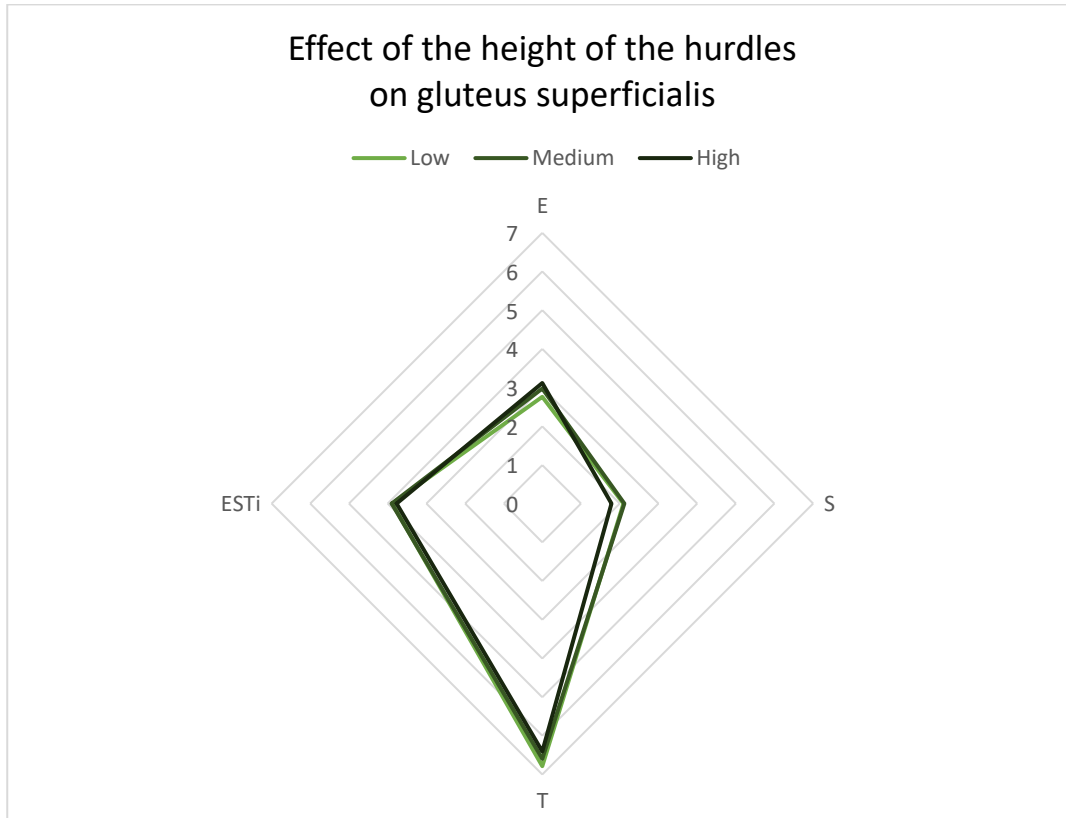


Figure 8: Radar diagram of the average recorded ESTi™-scores on gluteus superficialis on dogs (n = 14) when jumping hurdles with the heights of 45 cm (low = light green line), 55 cm (medium = green line) and 65 cm (high = dark green line) respectively. The axis from center and out shows the ESTi™-score. Data used for the radar diagram for gluteus superficialis can be found in Appendix L.

### 3.2.3 The descriptive data

No significant difference was found when clustering the dogs according to gender, height, age, experience, normal hurdle height in training and normal contact-method practiced on A-frame.

According to the weight of the dogs, the dogs weighing more than 21 kg ( $n=7$ ) had a significantly ( $p<0.05$ ) higher S-score for gluteus superficialis when jumping both medium ( $S=3.1$ ) and high ( $S=2.9$ ) hurdles compared to dogs weighing 21 kg or below ( $n=7$ ) (medium and high  $S=1.2$ ,  $S=0.7$  respectively). There was no significant difference when jumping the low hurdles. Dogs weighing more than 21 kg recruited less muscle fibers in gluteus superficialis compared to dogs weighing 21 kg or less. No significant difference was found in triceps.

According to training level the dogs with 1 hour of training per week or more ( $n=6$ ) had a significantly ( $p<0.05$ ) higher E-score for triceps brachii when jumping both medium ( $E=1.9$ ) and high ( $E=2.4$ ) hurdles compared to dogs with less than 1 hour of training per week ( $n=8$ ) (medium and high  $E=0.7$ ,  $E=0.8$  respectively). The ESTi-score was also significantly ( $p<0.05$ ) higher in triceps brachii for the dogs with 1 hour of training per week or more when jumping medium hurdles (ESTi=2.4), compared to dogs with less than 1 hour of training per week (ESTi=1.2). There was no significant difference when jumping the low hurdles. Dogs with 1 hour of training per week or more used triceps brachii more effective and coordinated when jumping medium and high hurdles compared to dogs with less than 1 hour of training per week. No significant difference was found in gluteus superficialis. See appendix L for data.

### 3.3 The A-frame -the effect of the height

#### 3.3.1 Triceps brachii

When testing the first run against the last of the three runs for each height no significant difference was found except for the T-score when running up the high A-frame. The mean T-score for the first run (T=7.7) was higher than the mean T-score for the last run (T=6.2) with a p-value of 0.012. The dogs had a significantly higher firing rate for triceps brachii in the last run when running up the high A-frame, indicating signs of fatigue.

##### 3.3.1.1 Running up the A-frame

No significant difference was found in the ESTi™-scores for triceps brachii when comparing low, medium and high positioning of the A-frame. Neither when all dogs were clustered together (n=14, figure 9) or when they were divided in two clusters with dogs practicing 2o2o-contact method (n=11, figure 10) and running contact method (n=3, figure 11) respectively (appendix M).

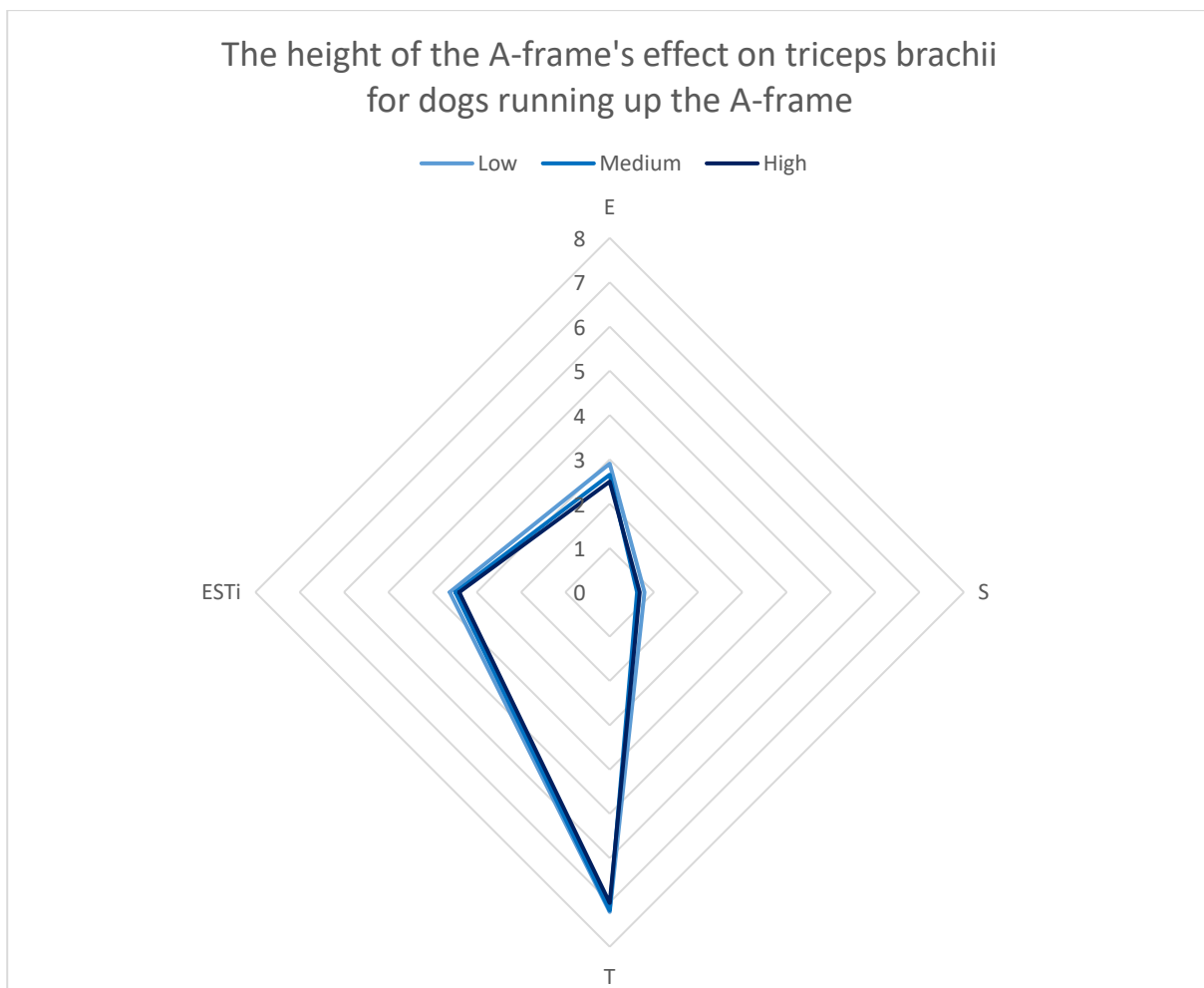


Figure 9: Radar diagram of the average recorded ESTi™-scores on triceps brachii on dogs (n = 14) running down the A-frame. The dogs ran the A-frame in three heights 118 cm (low = light blue line), 148 cm (medium = blue line) and 167 cm (high = dark blue line) respectively. The axis from center and out shows the ESTi™-score. Data used for the radar diagram for triceps brachii can be found in Appendix M.

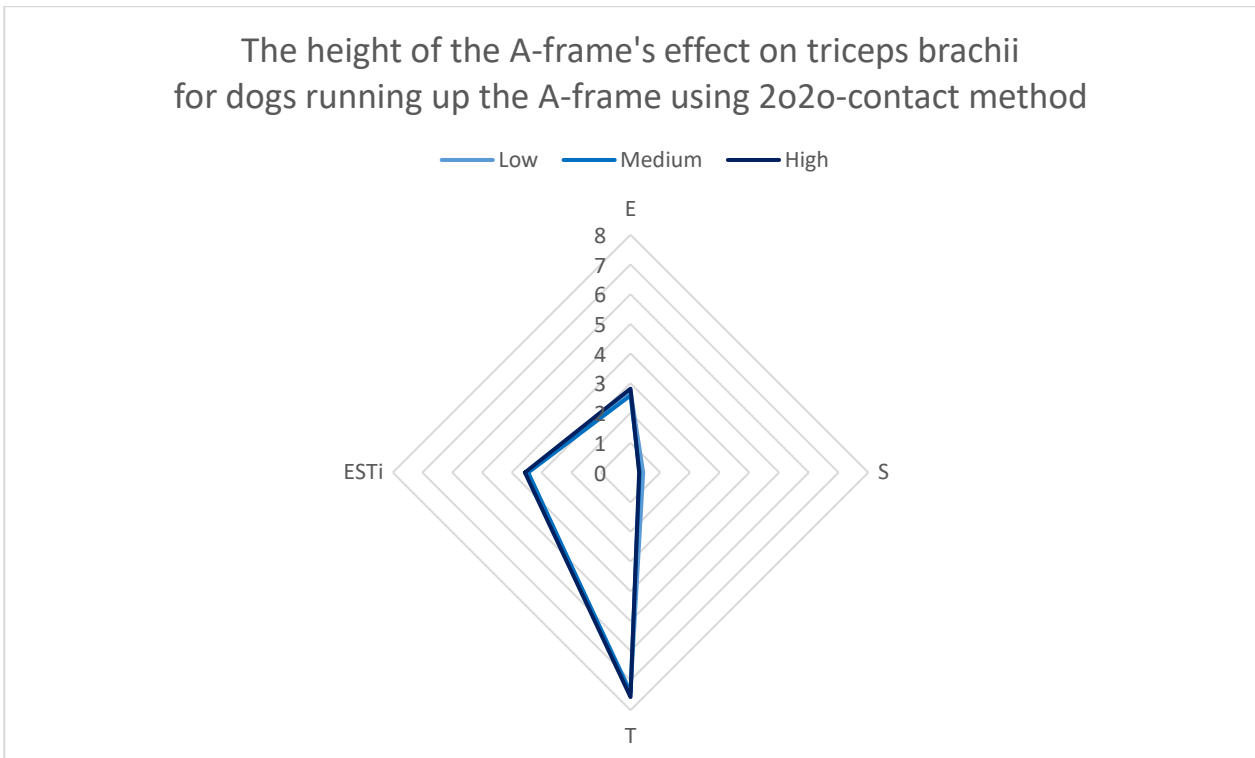


Figure 10: Radar diagram of the average recorded ESTi™-scores on triceps brachii on dogs (n = 11) running up the A-frame using 2o2o-contact method. The dogs ran the A-frame in three heights 118 cm (low = light blue line), 148 cm (medium = blue line) and 167 cm (high = dark blue line) respectively. The axis from center and out shows the ESTi™-score. Data used for the radar diagram for triceps brachii can be found in Appendix M.

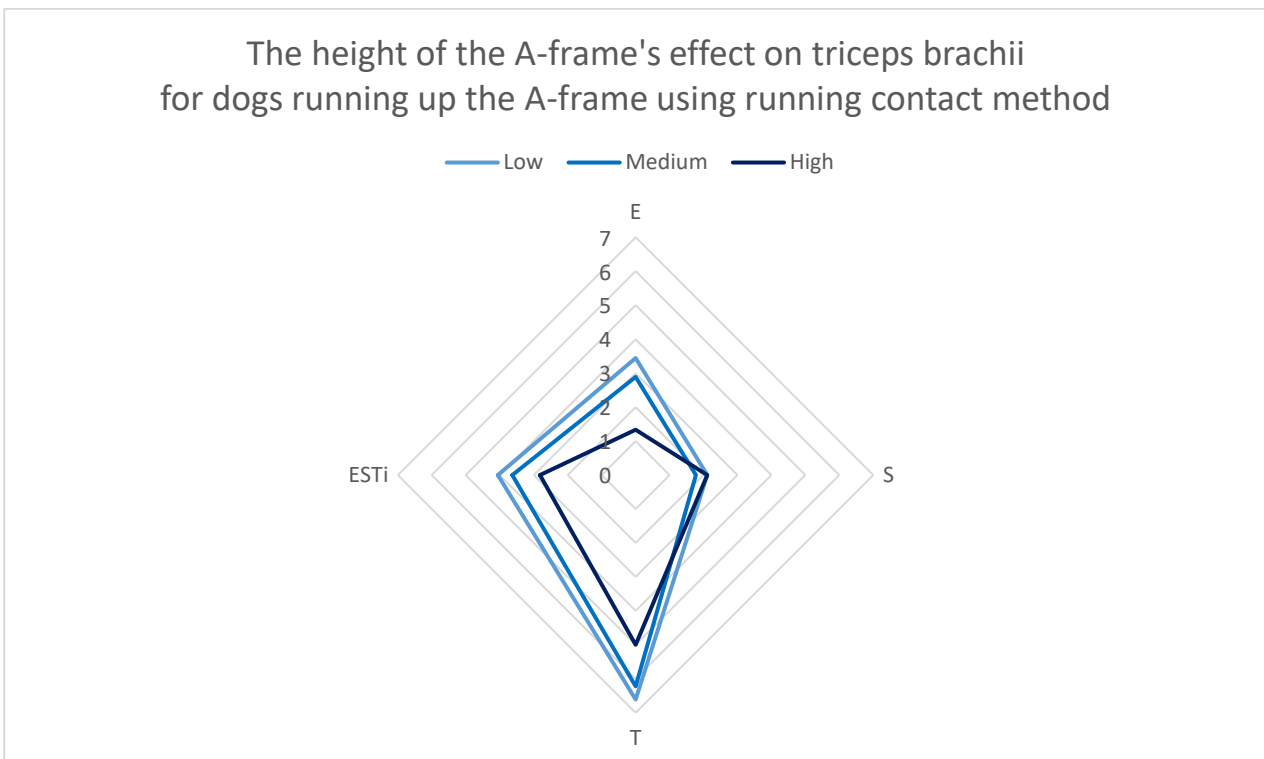


Figure 11: Radar diagram of the average recorded ESTi™-scores on triceps brachii on dogs (n = 3) running up the A-frame using running contact method. The dogs ran the A-frame in three heights 118 cm (low = light blue line), 148 cm (medium = blue line) and 167 cm (high = dark blue line) respectively. The axis from center and out shows the ESTi™-score. Data used for the radar diagram for triceps brachii can be found in Appendix M.

### 3.3.1.2 Running down the A-frame

No significant difference was found in the ESTi™-scores for triceps brachii when the dogs were clustered in the two groups with dogs practicing 2o2o-contact method (n=11, figure 13) and running contact method (n=3, figure 14) respectively. When all dogs were clustered together a significant difference (p<0.05) was found for the E-score when comparing low (E=3.9) with high (E=4.8) (n=14, figure 12). This means that, no matter the contact method used, the dogs used triceps brachii more efficient and coordinated when running down the high A-frames compared to low.

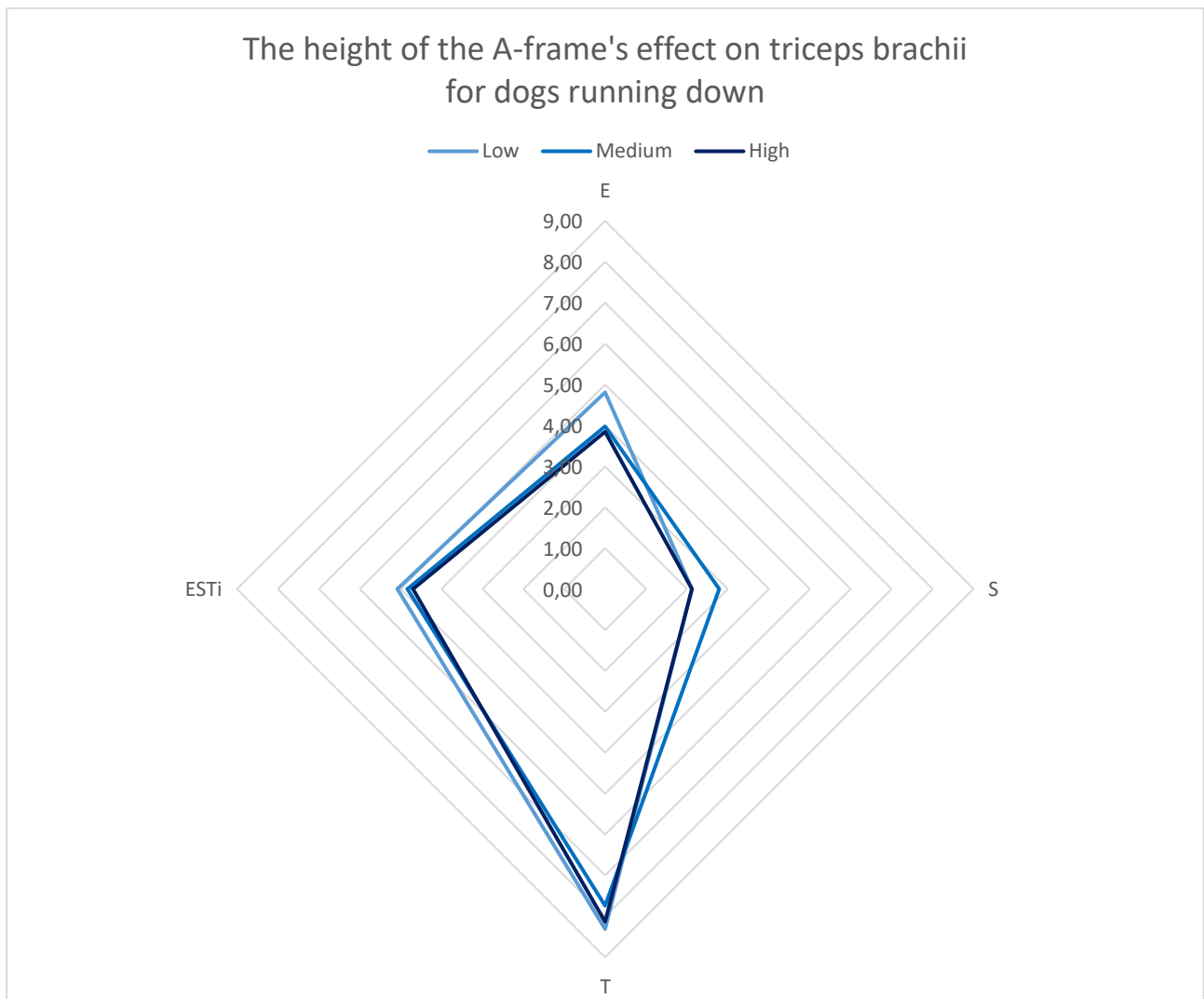


Figure 12: Radar diagram of the average recorded ESTi™-scores on triceps brachii on dogs (n = 14) running down the A-frame. The dogs ran the A-frame in three heights 118 cm (low = light blue line), 148 cm (medium = blue line) and 167 cm (high = dark blue line) respectively. The axis from center and out shows the ESTi™-score. Data used for the radar diagram for triceps brachii can be found in Appendix M.



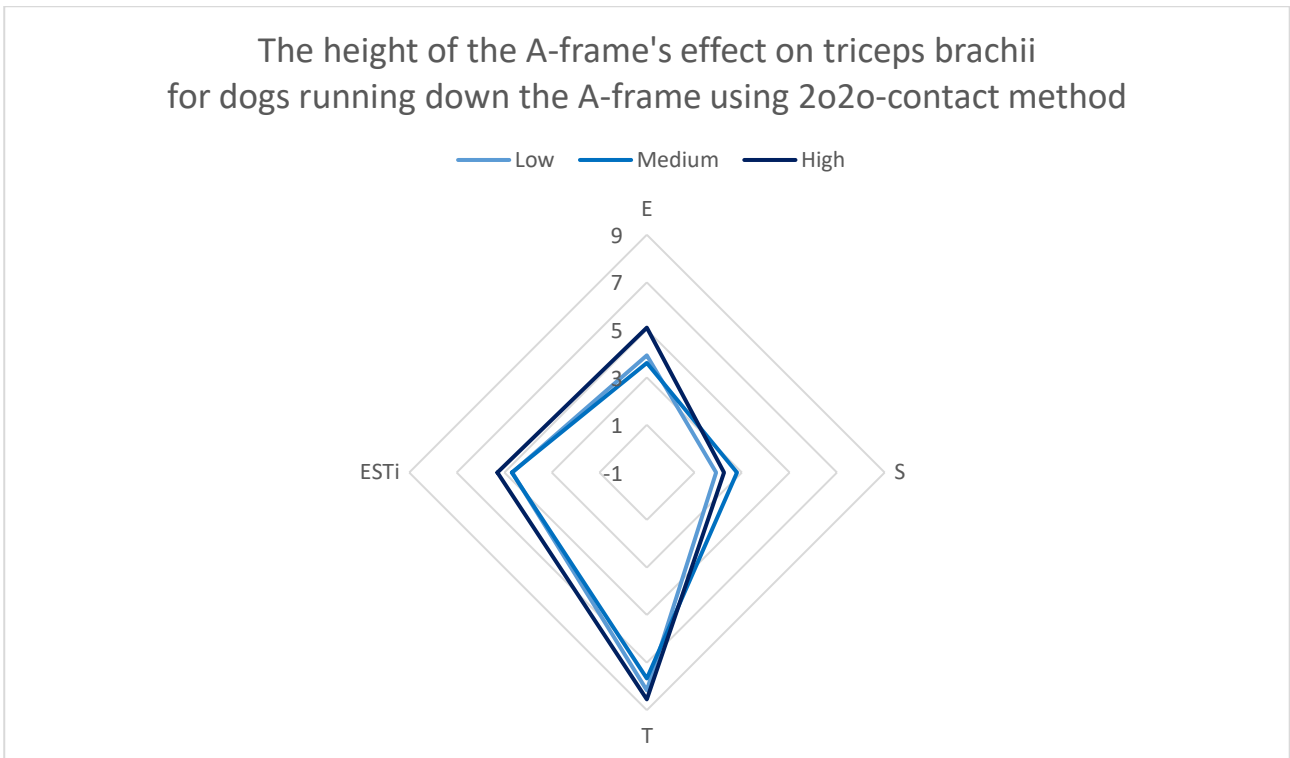


Figure 13: Radar diagram of the average recorded ESTi™-scores on triceps brachii on dogs (n = 11) running down the A-frame using 2o2o-contact method. The dogs ran the A-frame in three heights 118 cm (low = light blue line), 148 cm (medium = blue line) and 167 cm (high = dark blue line) respectively. The axis from center and out shows the ESTi™-score. Data used for the radar diagram for triceps brachii can be found in Appendix M.

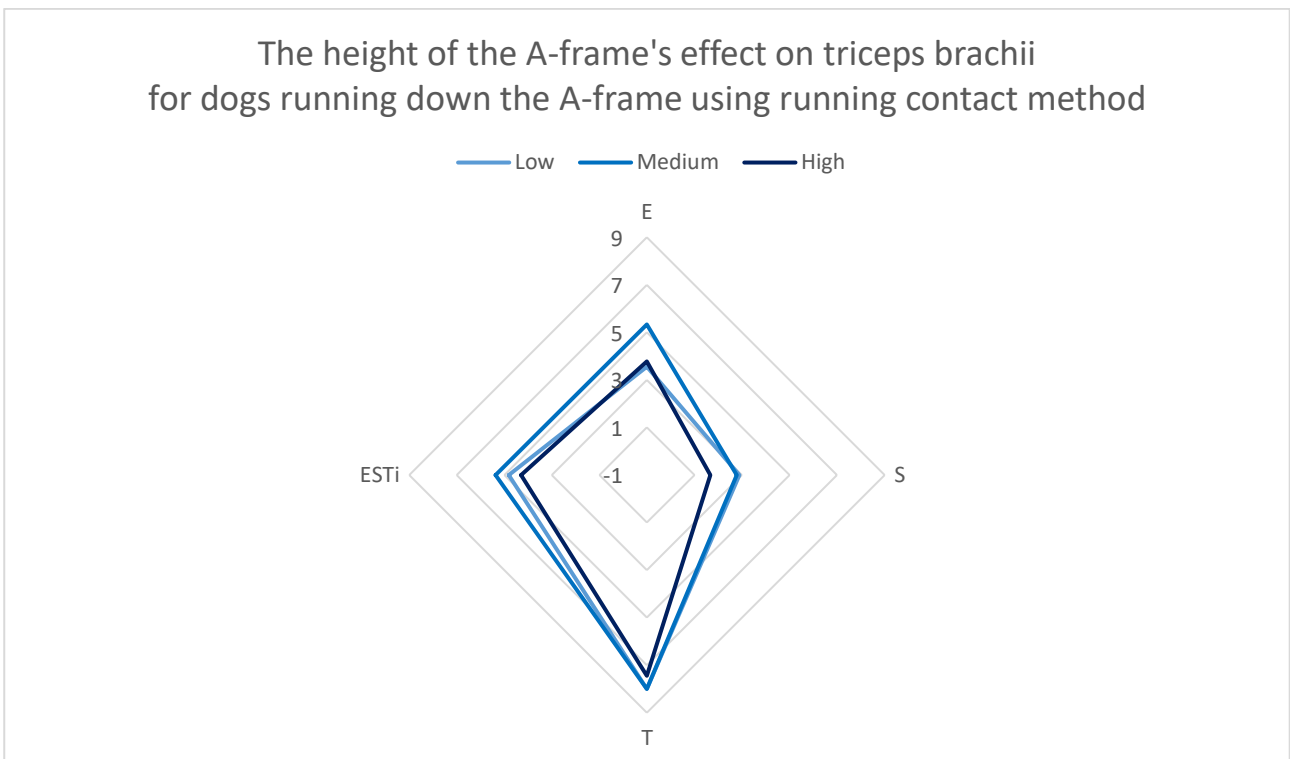


Figure 14: Radar diagram of the average recorded ESTi™-scores on triceps brachii on dogs (n = 3) running down the A-frame using running contact method. The dogs ran the A-frame in three heights 118 cm (low = light blue line), 148 cm (medium = blue line) and 167 cm (high = dark blue line) respectively. The axis from center and out shows the ESTi™-score. Data used for the radar diagram for triceps brachii can be found in Appendix M.

### 3.3.2 Gluteus superficialis

When testing the first run against the last of the three runs for each height no significant difference was found when running up the A-frame. Meanwhile a significant difference was found for the S-, T-, and ESTi-score when running down the medium A-frame. The mean S-, T-, and ESTi-score for the first run (S=6.3, T=7.3, ESTi=6.2) was higher than the last run (S=5.1, T=6.2, ESTi=5.2) with a p-value of 0.036, 0.045, 0.009 respectively. The dogs had a significantly higher firing rate and recruited more muscle fibers in triceps brachii in the last run when running down the medium A-frame, indicating signs of fatigue.

#### 3.3.2.1 *Running up the A-frame*

No significant difference was found in the ESTi<sup>TM</sup>-scores for gluteus superficialis when all dogs were clustered together (n=14, figure 15) or in a cluster with merely dogs practicing running contact method (n=3, figure 17). For dogs practicing 2o2o-contact method (n=11, figure 16) a significant difference (p<0.05) were found in the S-score from low (S=2.45) to high (S=2.12) A-frame. This is also illustrated in figure 16, meaning the dogs recruit more muscle fibers in gluteus superficialis when running up the high A-frame and practicing 2o2o-contact method compared to the low A-frame.

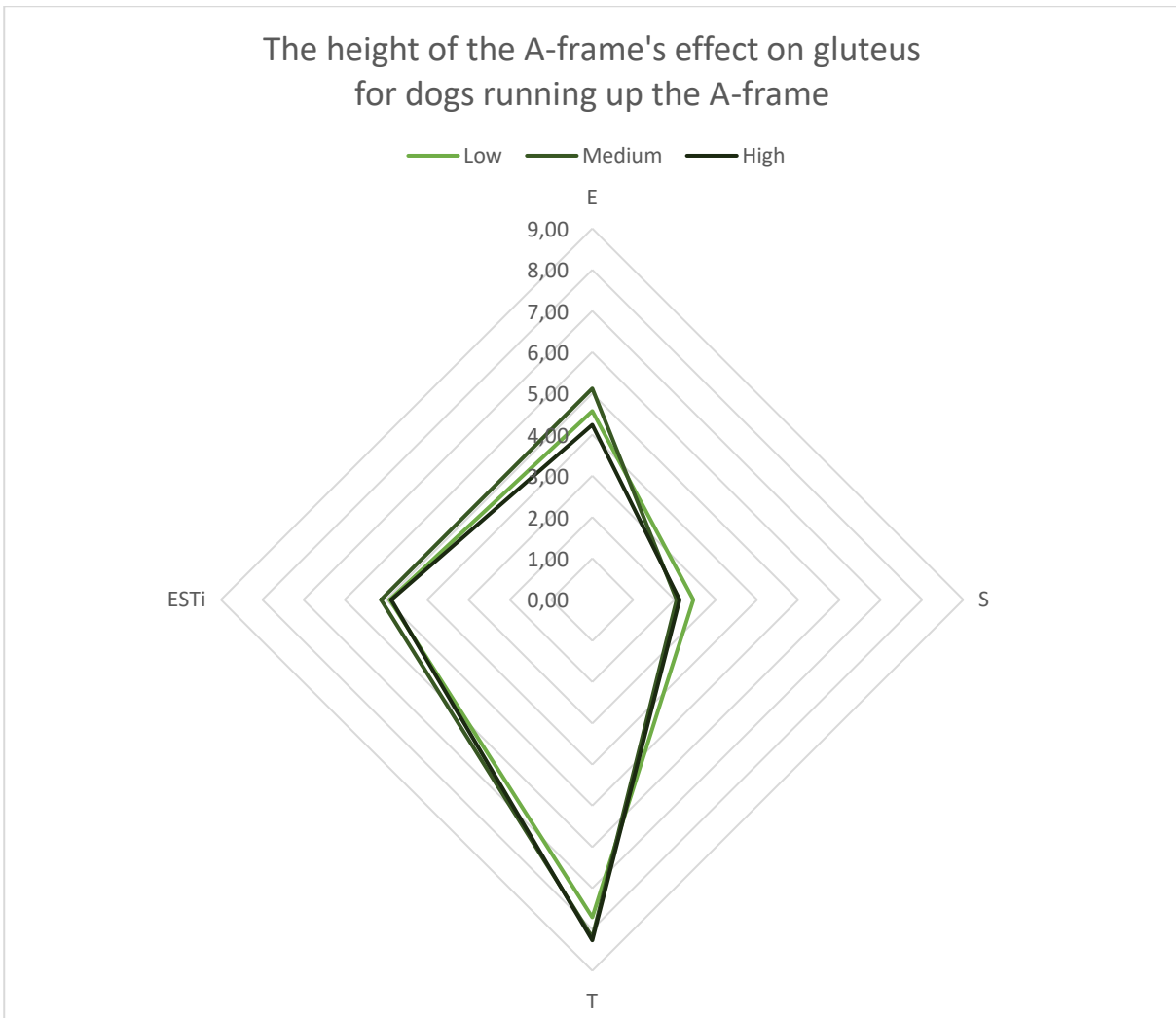


Figure 15: Radar diagram of the average recorded ESTi™-scores on gluteus superficialis on dogs (n = 14) running up the A-frame. The dogs ran the A-frame in three heights 118 cm (low = light green line), 148 cm (medium = green line) and 167 cm (high = dark green line) respectively. The axis from center and out shows the ESTi™-score. Data used for the radar diagram for gluteus superficialis can be found in Appendix M.

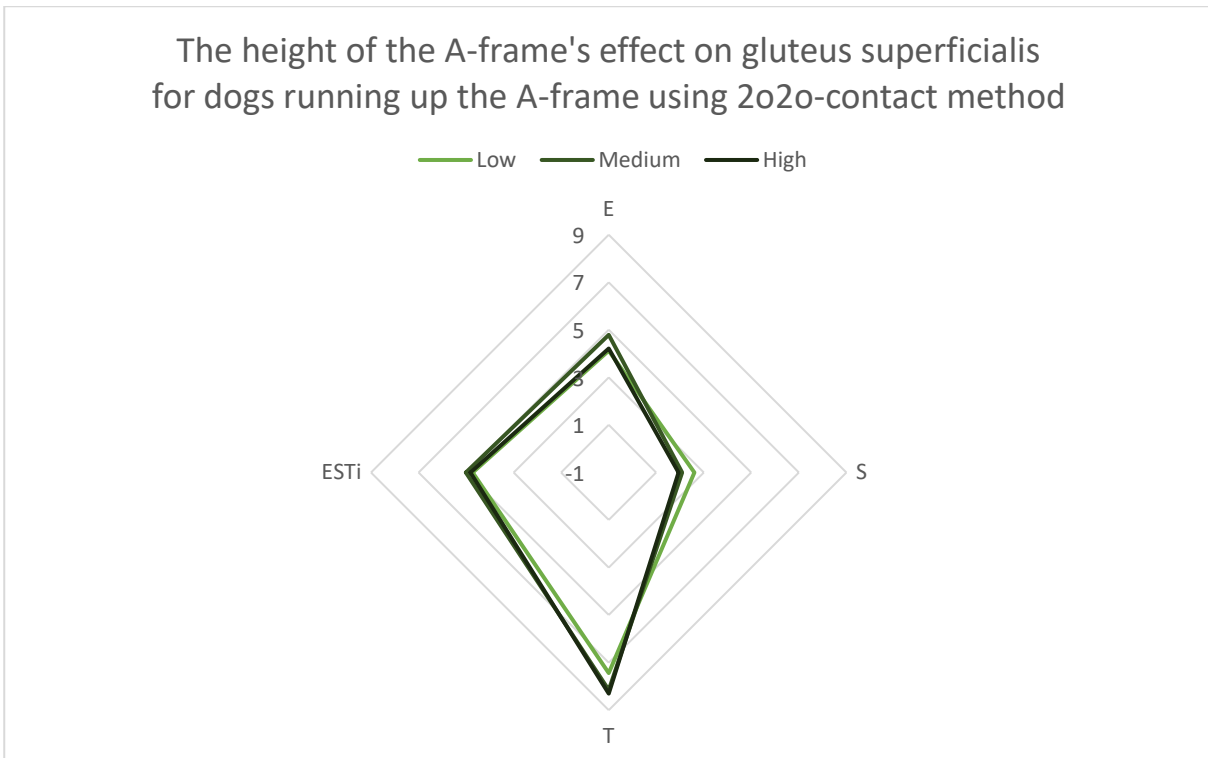


Figure 16: Radar diagram of the average recorded ESTi™-scores on gluteus superficialis on dogs (n = 3) running up the A-frame using 2o2o-contact method. The dogs ran the A-frame in three heights 118 cm (low = light green line), 148 cm (medium = green line) and 167 cm (high = dark green line) respectively. The axis from center and out shows the ESTi™-score. Data used for the radar diagram for gluteus superficialis can be found in Appendix M.

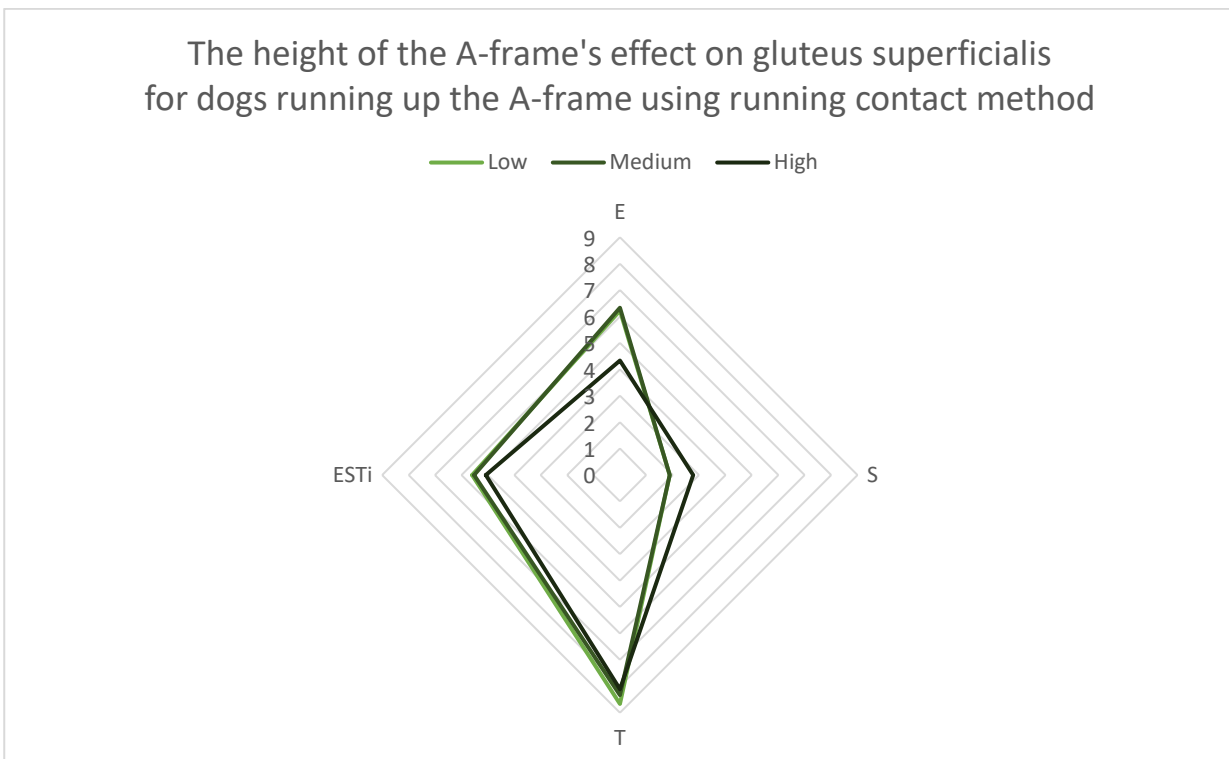


Figure 17: Radar diagram of the average recorded ESTi™-scores on gluteus superficialis on dogs (n = 3) running up the A-frame using running contact method. The dogs ran the A-frame in three heights 118 cm (low = light green line), 148 cm (medium = green line) and 167 cm (high = dark green line) respectively. The axis from center and out shows the ESTi™-score. Data used for the radar diagram for gluteus superficialis can be found in Appendix M.

### 3.3.2.2 Running down the A-frame

No significant difference was found in the ESTi™-scores for gluteus superficialis when comparing low, medium and high hurdles. Neither when all dogs were clustered together (n=14, figure 18) or when they were divided in two clusters with dogs practicing 2o2o-contact method (n=11, figure 19) and running contact method (n=3, figure 20) respectively.

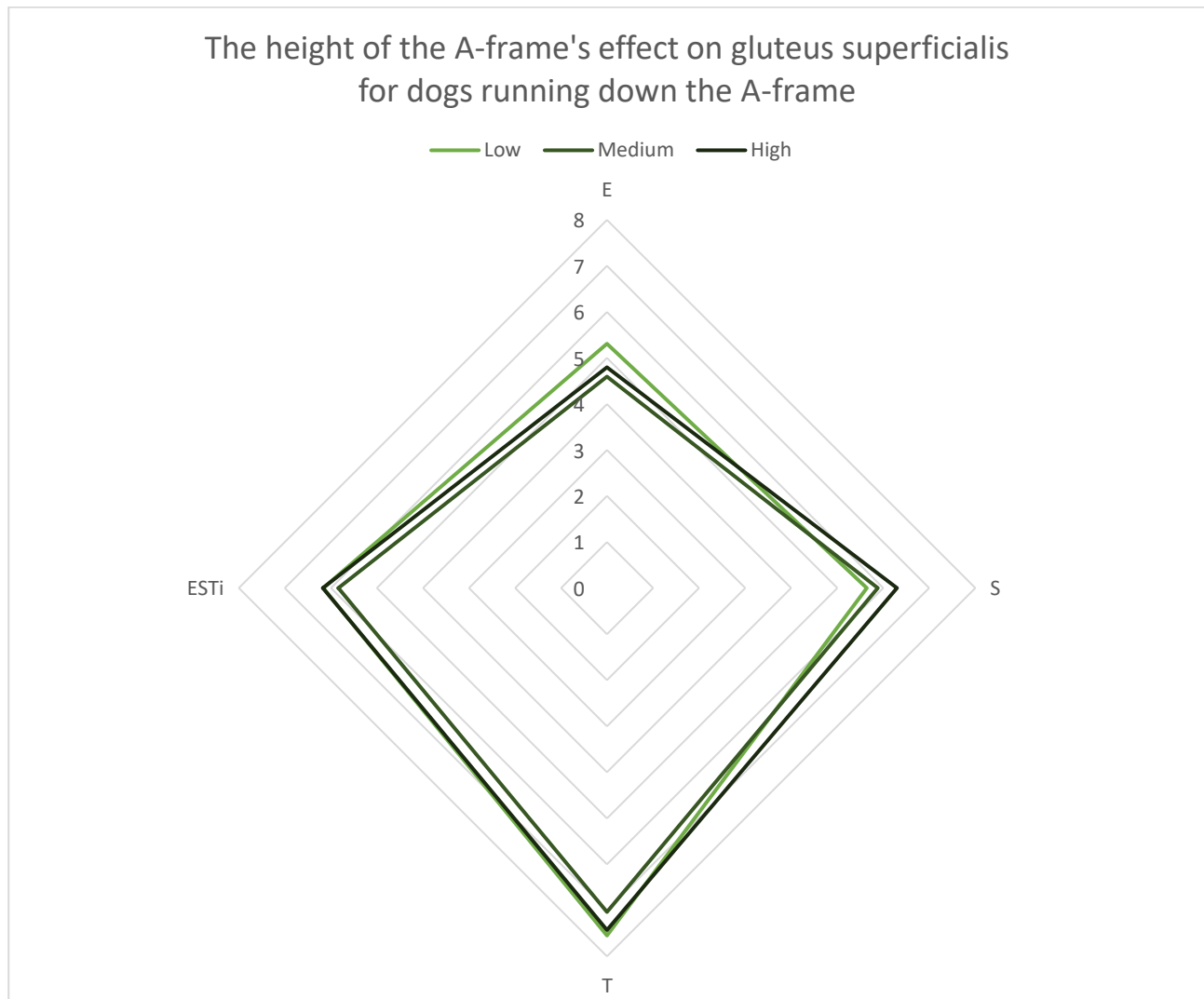


Figure 18: Radar diagram of the average recorded ESTi™-scores on gluteus superficialis on dogs (n = 14) running down the A-frame. The dogs ran the A-frame in three heights 118 cm (low = light green line), 148 cm (medium = green line) and 167 cm (high = dark green line) respectively. The axis from center and out shows the ESTi™-score. Data used for the radar diagram for gluteus superficialis can be found in Appendix M.

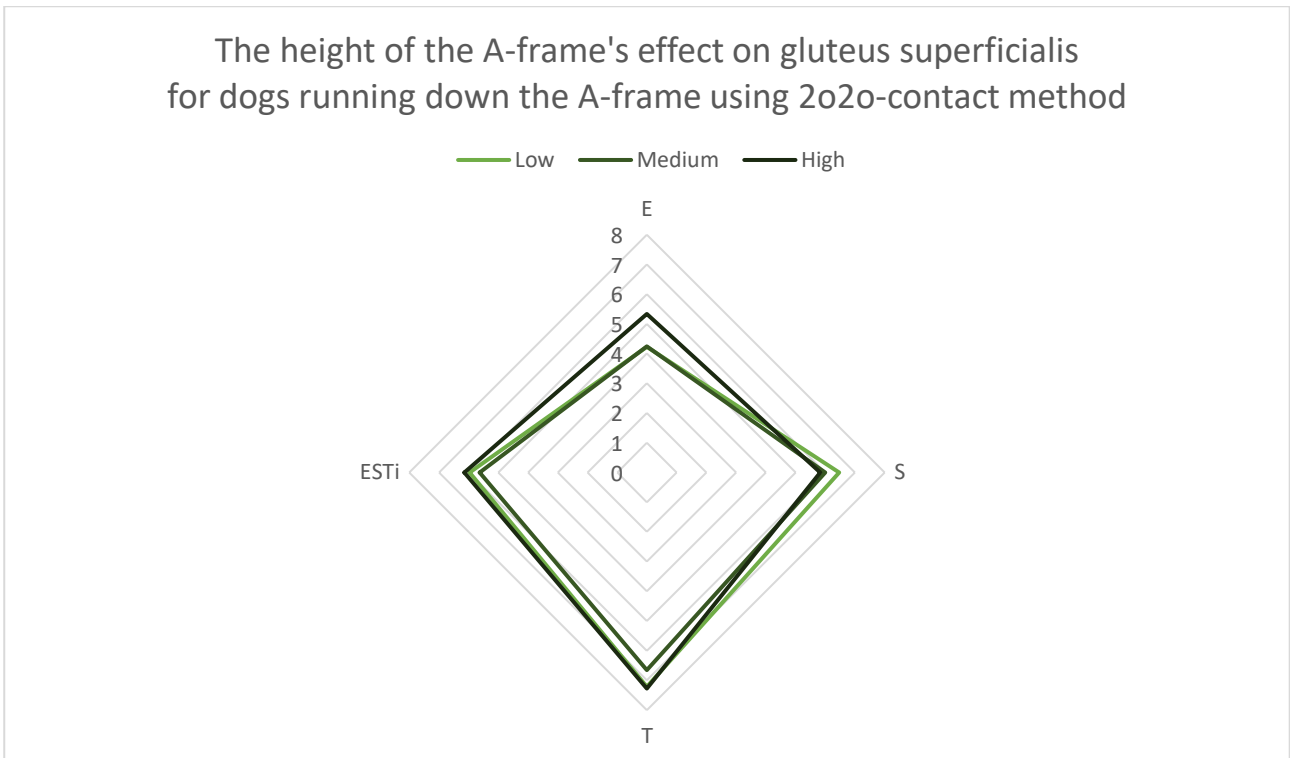


Figure 19: Radar diagram of the average recorded ESTi™-scores on gluteus superficialis on dogs (n = 3) running down the A-frame using 2o2o-contact method. The dogs ran the A-frame in three heights 118 cm (low = light green line), 148 cm (medium = green line) and 167 cm (high = dark green line) respectively. The axis from center and out shows the ESTi™-score. Data used for the radar diagram for gluteus superficialis can be found in Appendix M.

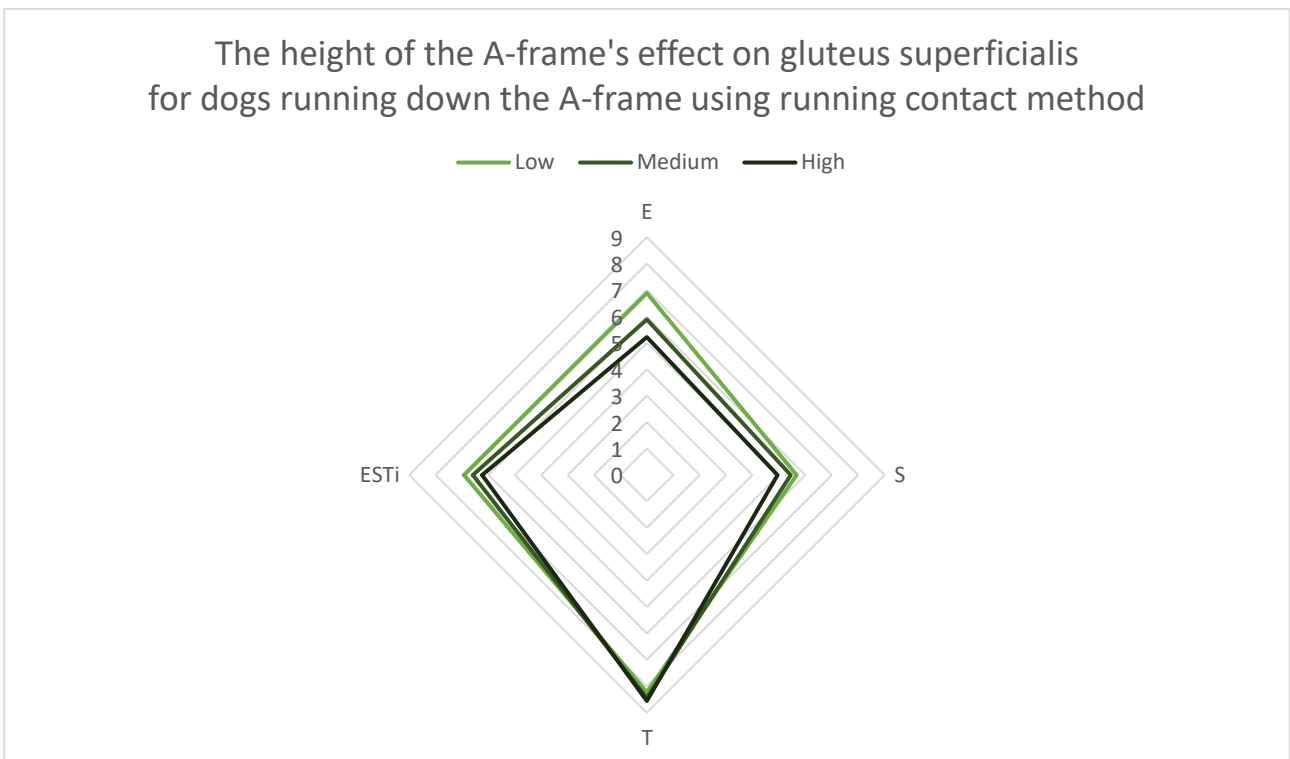


Figure 20: Radar diagram of the average recorded ESTi™-scores on gluteus superficialis on dogs (n = 3) running down the A-frame using running contact method. The dogs ran the A-frame in three heights 118 cm (low = light green line), 148 cm (medium = green line) and 167 cm (high = dark green line) respectively. The axis from center and out shows the ESTi™-score. Data used for the radar diagram for gluteus superficialis can be found in Appendix M.

### 3.3.3 The descriptive data

No significant difference was found when clustering the dogs according to weight and normal hurdle height in training. Only few significant differences with no obvious connection was found when clustering according to gender, age, experience and athletic score.

According to the height of the dogs, the dogs higher than 55 cm ( $n=7$ ) had a significantly ( $p<0.05$ ) lower T-score for triceps brachii when running up the A-frame in both medium ( $T=6.38$ ) and high ( $T=6.2$ ) position compared to dogs with a height of 55 cm or below ( $n=7$ ) (medium and high  $T=8.0$ ,  $T=7.8$  respectively). There was no significant difference when running the low A-frame. No significant difference was found in gluteus superficialis when running up the A-frame, and no significant difference was found in neither muscle when running down the A-frame.

According to training level the dogs with 1 hour of training per week or more ( $n=6$ ) had a significantly ( $p<0.05$ ) higher ESTi-score for triceps brachii when running up the A-frame in both low ( $E=4.6$ ) medium ( $E=4.3$ ) and high ( $3.9$ ) position compared to dogs with less than 1 hour of training per week ( $n=8$ ) (low, medium and high  $E=2.9$ ,  $E=2.9$ ,  $E=3.0$  respectively). Dogs with 1 hour of training per week or more had a significantly ( $p<0.05$ ) higher E-score for triceps brachii when running up the A-frame in both low ( $E=4.0$ ) and medium ( $E=3.8$ ) position compared to dogs with less than 1 hour of training per week (low and medium  $E=2.1$ ,  $E=1.8$  respectively). The S-score was also significantly ( $p<0.05$ ) higher in triceps brachii for the dogs with 1 hour of training per week or more when running up the high A-frame in medium ( $S=1.3$ ) and high ( $S=1.4$ ) position, compared to dogs with less than 1 hour of training per week (medium and high  $S=0.1$ ,  $S=0.1$  respectively). Dogs with 1 hour of training per week or more used triceps brachii more effective and coordinated when running up the low and medium A-frame and recruited less muscle fibers when running up the medium and high A-frame compared to dogs with less than 1 hour of training per week and were generally better at using their muscles when running all heights. No significant difference was found in gluteus superficialis.

When running down the A-frame the dogs with 1 hour of training per week or more had a significantly ( $p<0.05$ ) higher E-score for triceps brachii when running the A-frame in both low ( $E=5.1$ ) and high ( $E=6.3$ ) position. They also had a higher S-score for medium ( $S=4.4$ ) A-frame compared to dogs with less than 1 hour of training per week (low, medium and high  $E=2.9$ ,  $S=1.6$ ,  $E=3.7$  respectively). Dogs with 1 hour of training per week or more used triceps brachii more effective and coordinated when running down the low and high A-frame. They also recruited less muscle fibers when running down the medium A-frame compared to dogs with less than 1 hour of training per week. No significant difference was found in gluteus superficialis. See appendix M for data.



## 3.4 The A-frame - the effect of contact method

### 3.4.1 Triceps brachii

When the first run was tested against the last of the three runs no significant difference was found when practicing the unusual contact method.

#### 3.4.1.1 Running up the A-frame

Comparing figure 10 with figure 11 the dogs used to practice 2o2o-contact method had a significantly ( $p < 0.05$ ) higher T- and ESTi-score ( $T=7.6$ ,  $ESTi=3.6$ ) compared to dogs used to practice running contact method ( $T=5.0$ ,  $ESTi=2.8$ ) when running up the high A-frame (appendix M). Dogs used to practice 2o2o-contact method had a lower firing rate in triceps brachii compared to dogs used to practice running contact method and an overall improved use of triceps.

When making no difference of the dogs according to what they are used to practice the 2o2o-contact method had a significant higher E-score (2.7 vs 1.8) and ESTi-score (3.5 vs 3.1) for triceps brachii when running up the A-frame (table 10). The same was the case with the dogs used to practicing the 2o2o-contact method. For the dogs used to practice running contact method there were no difference when they shifted to 2o2o-contact method. Triceps brachii are used more efficient and coordinated when practicing 2o2o-contact method. This is also illustrated in figure 21.

Table 10: The average recorded ESTi™-scores of triceps brachii on dogs running up the A-frame practicing either 2o2o-contact method or running contact method. The dogs are listed both in one group and divided into groups in relation to what they normally practice as contact method. A T-test was used for data being normally distributed otherwise a Wilcoxon Signed-Ranks Test were performed and the p-value listed.

Clusters	Score	2o2o-contact method		Running contact method		Test	p-value
		Mean	SD	Mean	SD		
Both contact methods n=14	E	2,7	0,9	1,8	1,0	T-test	0,005
	S	0,7	1,4	0,7	1,7	Wilcoxon	0,779
	T	7,2	1,1	6,7	1,6	Wilcoxon	0,286
	ESTi	3,5	0,8	3,1	0,5	T-test	0,017
2o2o-contact method n=11	E	2,8	1,0	1,9	0,7	T-test	0,008
	S	0,3	0,3	0,3	0,5	Wilcoxon	0,753
	T	7,6	0,7	7,2	1,1	Wilcoxon	0,351
	ESTi	3,6	0,6	3,1	0,5	T-test	0,012
Running contact method n=3	E	2,4	0,8	1,3	2,0	T-test	0,388
	S	2,0	2,9	2,1	3,7	Wilcoxon	0,655
	T	5,8	1,6	5,0	2,2	T-test	0,732
	ESTi	3,4	1,5	2,8	0,3	Wilcoxon	0,593

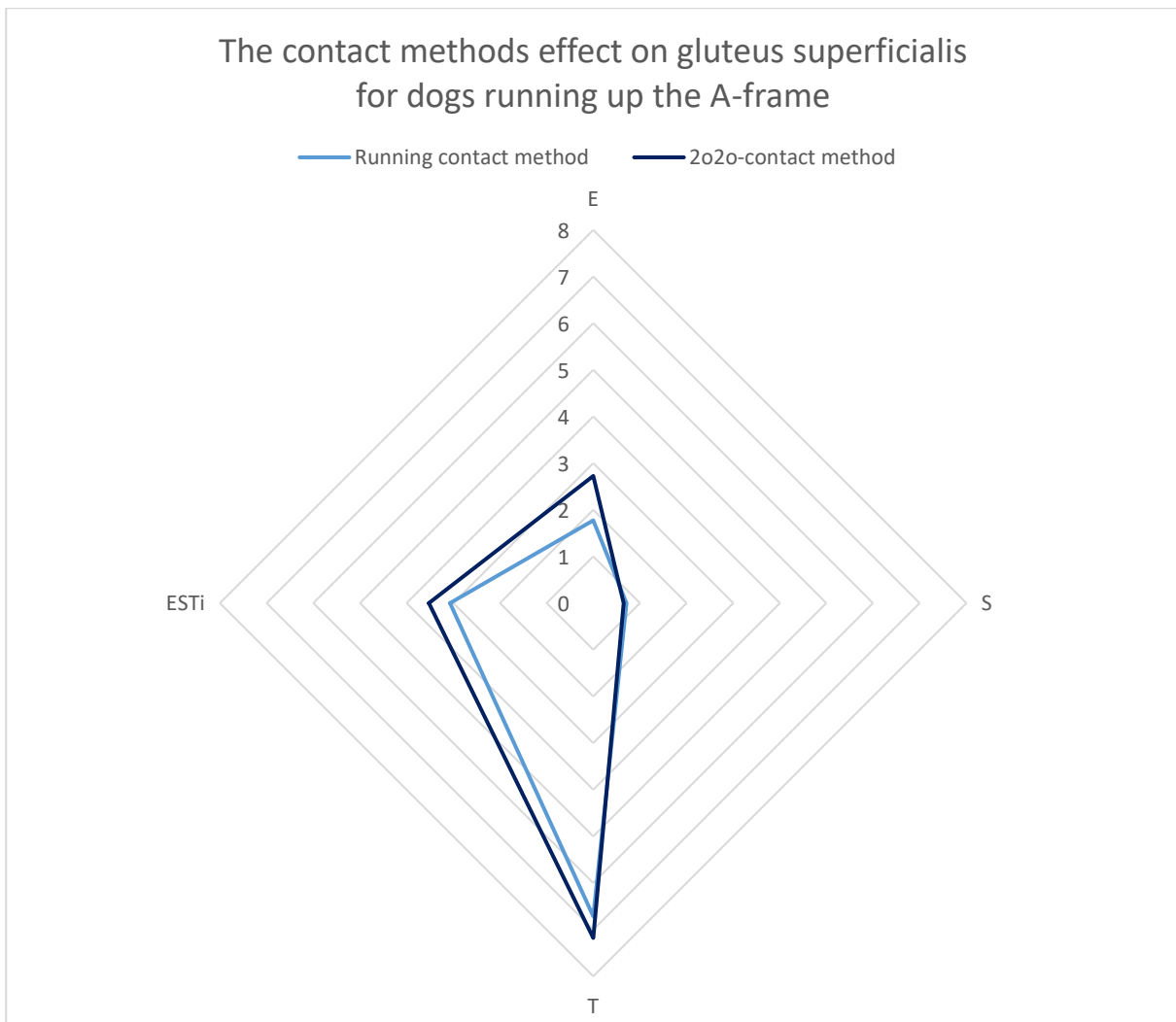


Figure 21: Radar diagram of the average recorded ESTi™-scores on triceps brachii on dogs (n = 14) running up the A-frame. The dogs ran the A-frame when practicing either running contact method (light blue line) or 2o2o-contact method (dark blue line). The axis from center and out shows the ESTi™-score. Data used for the radar diagram for triceps brachii can be found in Appendix M.

### 3.4.1.2 Running down the A-frame

There was no significant difference when comparing dogs used to practice 2o2o-contact method with dogs used to practice running contact method. Notice: it is results from figure 13 compared with results from figure 14.

When making no difference of the dogs according to what they are used to practice the 2o2o-contact method had a significant higher E-score (4.6 vs 2.8) and ESTi-score (5.1 vs 4.1) for triceps brachii when running down the A-frame (table 11, figure 22). The same was the case with the dogs used to practice the 2o2o-contact method, but they also had a higher T-score (8.5 vs 8.0). For the dogs used to practice running contact method there were no difference when they shifted to 2o2o-contact method. Triceps brachii are used more efficient and coordinated when practicing 2o2o-contact method. The dogs used to practice the 2o2o-contact method used triceps brachii with a lower firing rate compared to when they shifted to practicing the running contact method.

Table 11: The average recorded ESTi<sup>TM</sup>-scores of triceps brachii on dogs running down the A-frame practicing either 2o2o-contact method or running contact method. The dogs are listed both in one group and divided into groups in relation to what they normally practice as contact method. A T-test was used for data being normally distributed otherwise a Wilcoxon Signed-Ranks Test were performed and the p-value listed. Data used for the table can be found in Appendix M.

Clusters	Score	2o2o-contact method		Running contact method		Test	p-value
		Mean	SD	Mean	SD		
Both contact methods n=14	E	4,6	1,8	2,8	1,9	T-test	0,011
	S	2,7	1,9	1,7	1,5	T-test	0,209
	T	8,3	0,8	7,9	0,7	T-test	0,084
	ESTi	5,1	1,2	4,1	1,2	T-test	0,013
2o2o-contact method n=11	E	5,1	2,2	2,6	1,8	T-test	0,007
	S	2,2	1,5	1,7	1,5	Wilcoxon	0,285
	T	8,5	0,5	8,0	0,7	Wilcoxon	0,041
	ESTi	5,3	1,2	4,1	1,1	T-test	0,017
Running contact method n=3	E	4,0	0,6	3,8	2,3	Wilcoxon	0,886
	S	2,2	2,8	1,7	1,7	Wilcoxon	0,655
	T	7,3	0,9	7,4	0,8	T-test	0,868
	ESTi	4,5	1,2	4,3	1,5	T-test	0,285

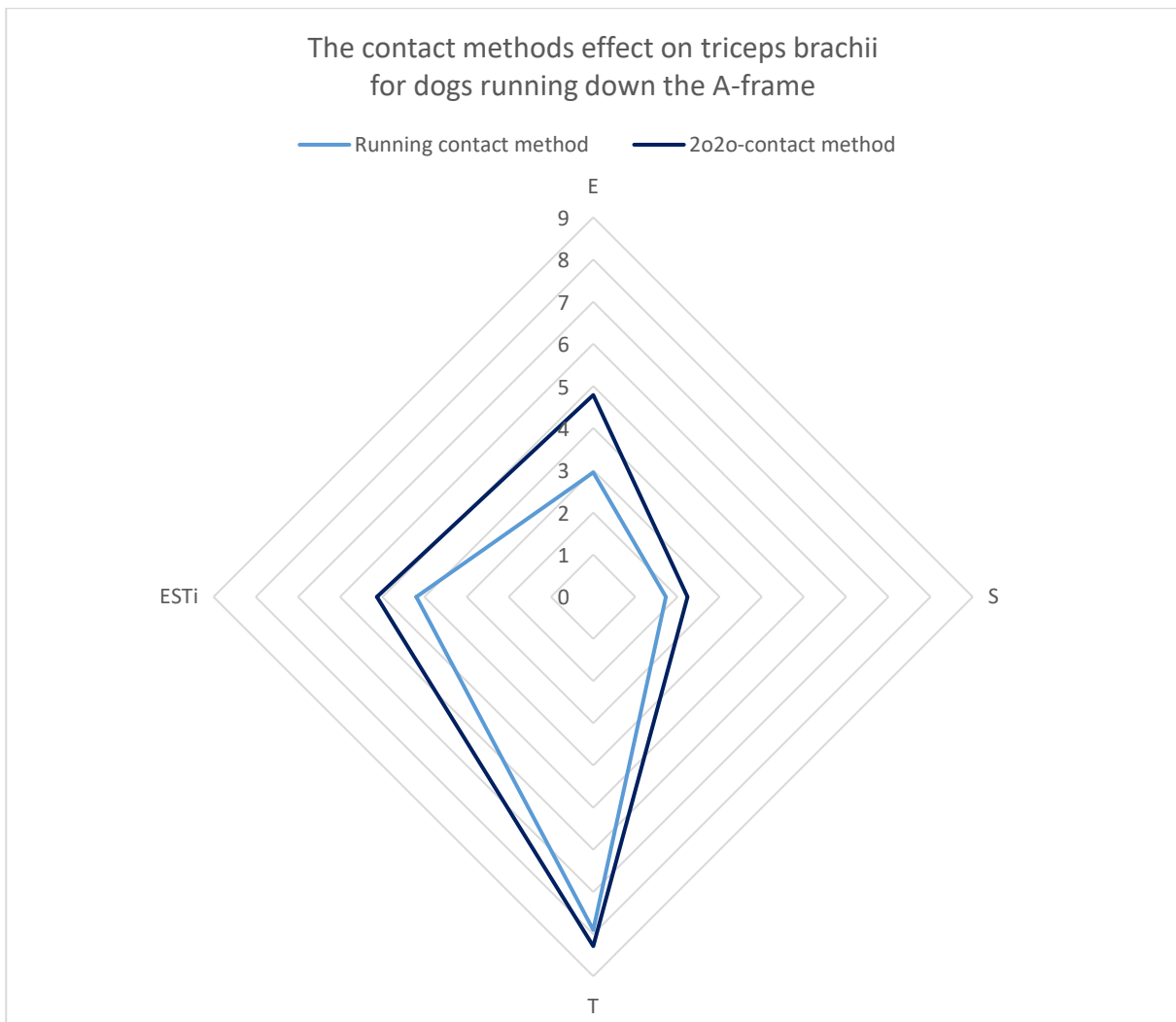


Figure 22: Radar diagram of the average recorded ESTi™-scores on triceps brachii on dogs (n = 14) running down the A-frame. The dogs ran the A-frame when practicing either running contact method (light blue line) or 2o2o-contact method (dark blue line). The axis from center and out shows the ESTi™-score. Data used for the radar diagram for triceps brachii can be found in Appendix M.

### 3.4.2 Gluteus superficialis

When testing the first run against the last of the three runs for both contact methods used no significant difference was found, indicating no signs of fatigue or improvement.

#### 3.4.2.1 Running up the A-frame

There was no significant difference when comparing dogs used to practice 2o2o-contact method with dogs used to practice running contact method. Notice: it is results from figure 16 compared with results from figure 17.

No significant difference was found in the ESTi<sup>TM</sup>-scores for triceps brachii when comparing the two contact methods (table 12) when running up the A-frame. Neither when all dogs were clustered together (n=14) or when they were divided in two clusters with dogs used to practicing 2o2o-contact method (n=11) and running contact method (n=3) respectively. Figure 23 illustrate the same result with only small differences in the ESTi<sup>TM</sup>-score to be seen.

Table 12: The average recorded ESTi<sup>TM</sup>-scores of gluteus superficialis on dogs running up the A-frame practicing either 2o2o-contact method or running contact method. The dogs are listed both in one group and divided into groups in relation to what they normally practice as contact method. A T-test was used for data being normally distributed otherwise a Wilcoxon Signed-Ranks Test were performed and the p-value listed. Data used for the table can be found in Appendix M.

Clusters	Score	2o2o-contact method		Running contact method		Test	p-value
		Mean	SD	Mean	SD		
Both contact methods n=14	E	4,5	1,5	3,9	1,9	T-test	0,280
	S	1,9	1,7	1,9	1,9	Wilcoxon	0,875
	T	8,2	0,6	7,8	1,2	Wilcoxon	0,551
	ESTi	4,8	0,8	4,6	0,9	T-test	0,274
2o2o-contact method n=11	E	4,2	1,4	3,8	1,7	T-test	0,442
	S	1,9	1,9	1,7	1,8	Wilcoxon	0,477
	T	8,3	0,3	7,8	1,4	Wilcoxon	0,307
	ESTi	4,8	0,8	4,4	0,9	T-test	0,205
Running contact method n=3	E	5,6	1,6	4,3	2,9	T-test	0,556
	S	1,6	1,1	2,8	2,5	T-test	0,267
	T	7,8	1,1	8,1	0,2	Wilcoxon	0,423
	ESTi	5,0	0,9	5,1	0,7	T-test	0,852

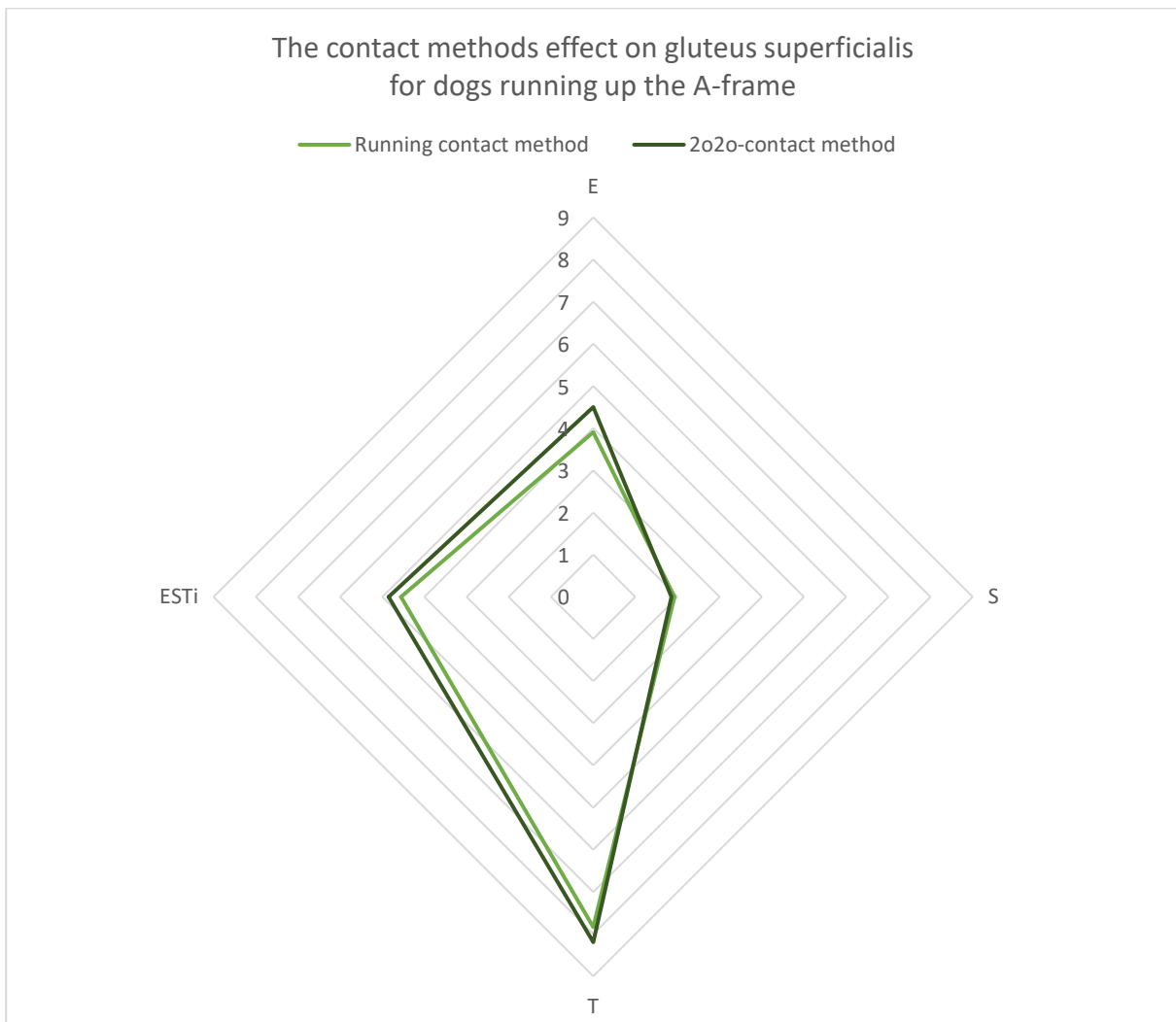


Figure 23: Radar diagram of the average recorded ESTi™-scores on gluteus superficialis on dogs (n = 14) running up the A-frame. The dogs ran the A-frame when practicing either running contact method (light green line) or 2o2o-contact method (dark green line). The axis from center and out shows the ESTi™-score. Data used for the radar diagram for gluteus superficialis can be found in Appendix M.



### 3.4.2.2 Running down the A-frame

There was no significant difference when comparing dogs used to practice 2o2o-contact method with dogs used to practice running contact method. Notice: it is results from figure 19 compared with results from figure 20.

When making no difference of the dogs according to what they are used to practice the 2o2o-contact method had a significant higher E-score for gluteus superficialis when running down the A-frame (table 13). The ESTi-score was significantly higher for the dogs used to practice the 2o2o-contact method. Meanwhile, for the dogs used to practice running contact method there were no difference when they shifted to 2o2o-contact method. Triceps brachii are used more efficient and coordinated when practicing 2o2o-contact method when considering all the dogs. For dogs used to practice the 2o2o-contact method, triceps brachii was overall used better when practicing the 2o2o-contact method. This is also illustrated in figure 24 with a visibly higher E and ESTi-score when practicing 2o2o-contact method.

Table 13: The average recorded ESTi™-scores of gluteus superficialis on dogs running down the A-frame practicing either 2o2o-contact method or running contact method. The dogs are listed both in one group and divided into groups in relation to what they normally practice as contact method. A T-test was used for data being normally distributed otherwise a Wilcoxon Signed-Ranks Test were performed and the p-value listed. Data used for the table can be found in Appendix M.

Clusters	Score	2o2o-contact method		Running contact method		Test	p-value
		Mean	SD	Mean	SD		
Both contact methods n=14	E	5,5	2,1	4,0	2,1	T-test	0,045
	S	5,4	2,5	5,4	2,6	Wilcoxon	0,463
	T	7,5	1,7	7,6	1,3	Wilcoxon	0,875
	ESTi	6,1	1,1	5,6	0,9	T-test	0,051
2o2o-contact method n=11	E	5,3	2,2	3,6	1,9	T-test	0,062
	S	5,8	2,5	5,5	2,8	Wilcoxon	0,508
	T	7,3	1,9	7,3	1,3	Wilcoxon	0,657
	ESTi	6,1	1,1	5,5	1,0	T-test	0,030
Running contact method n=3	E	6,3	1,5	5,2	2,5	T-test	0,590
	S	3,9	2,3	4,9	2,2	T-test	0,710
	T	8,2	1,1	8,6	1,3	T-test	0,285
	ESTi	6,1	1,3	6,2	0,6	T-test	0,841

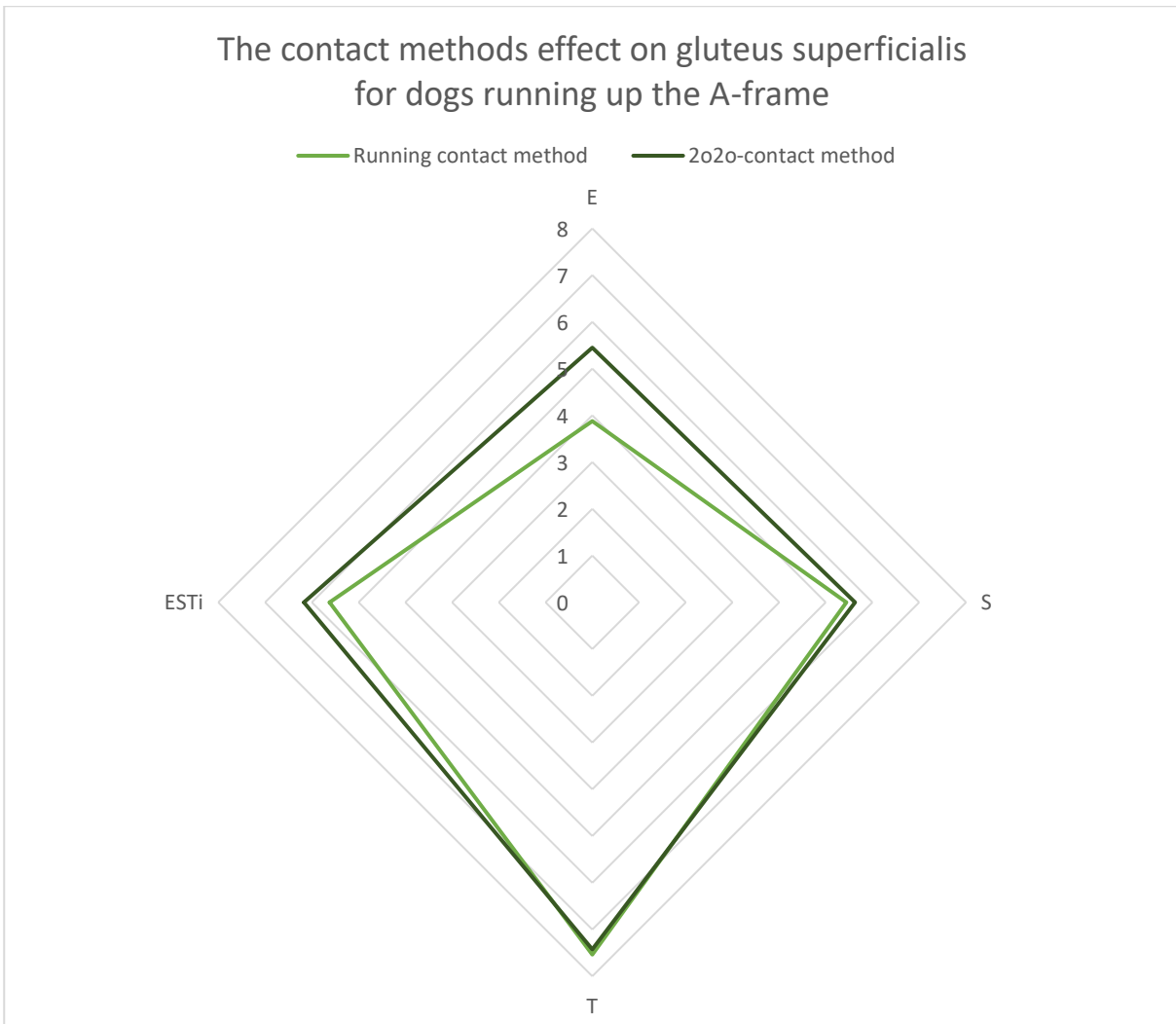


Figure 24: Radar diagram of the average recorded ESTi™-scores on gluteus superficialis on dogs (n = 14) running down the A-frame. The dogs ran the A-frame when practicing either running contact method (light green line) or 2o2o-contact method (dark green line). The axis from center and out shows the ESTi™-score. Data used for the radar diagram for gluteus superficialis can be found in Appendix M.

## 3.5 The cool-down exercises

### 3.5.1 Triceps brachii

No significant difference was found for triceps brachii when comparing the ESTi™-scores before and after cool-down exercises. This is also clearly illustrated in figure 25.

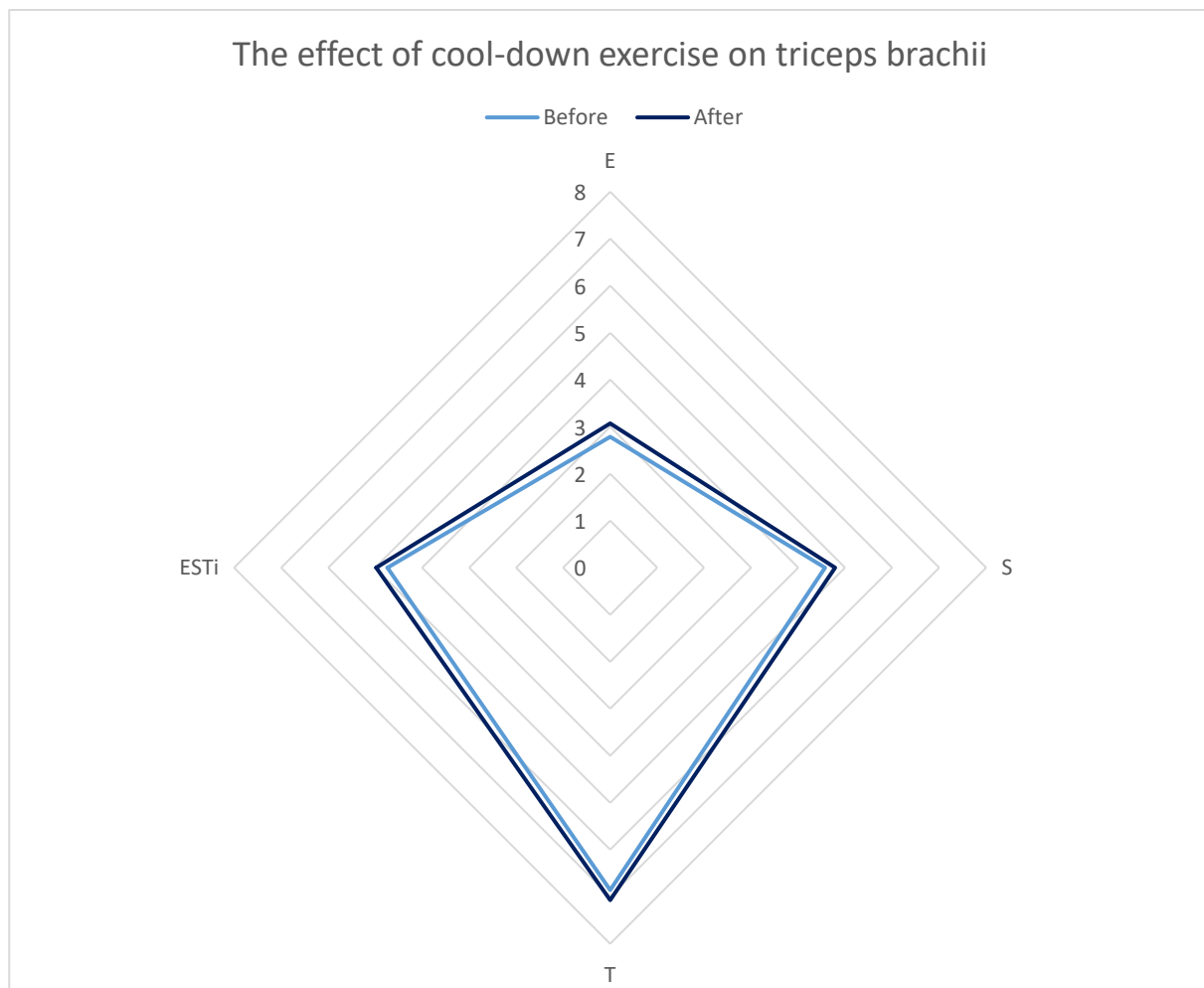


Figure 25: Radar diagram of the average recorded ESTi™-scores of triceps brachii on dogs (n = 14) before (light blue line) and after (dark blue) performing 15 minutes of cool-down exercises. The axis from center and out shows the ESTi™-score. Data used for the radar diagram for triceps brachii can be found in Appendix N.

### 3.5.2 Gluteus superficialis

No significant difference was found for gluteus superficialis when comparing the ESTi™-scores before and after cool-down exercises. This is also clearly illustrated in figure 26.

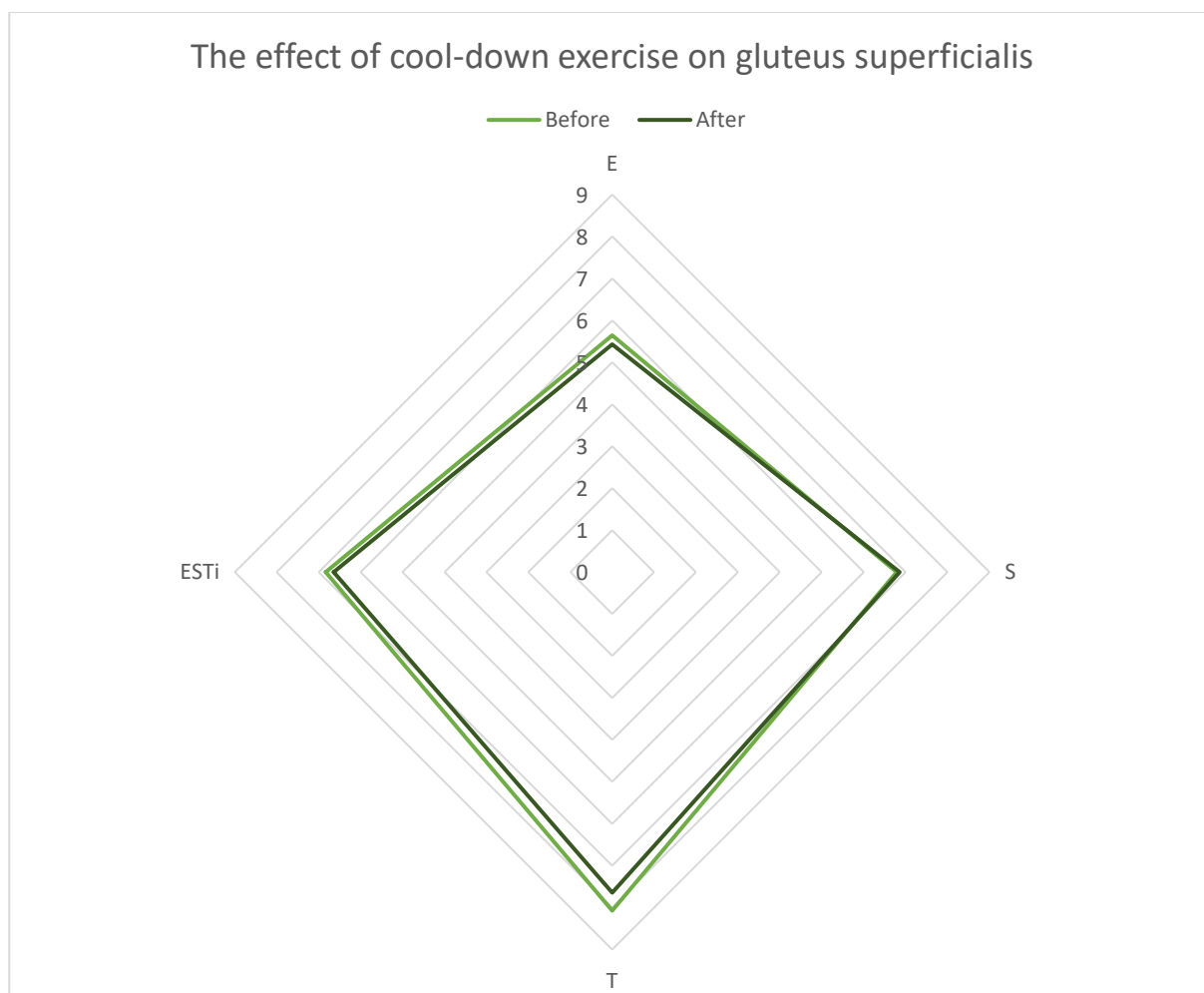


Figure 26: Radar diagram of the average recorded ESTi™-scores of gluteus superficialis on dogs (n = 14) before (light green line) and after (dark green line) performing 15 minutes of cool-down exercises. The axis from center and out shows the ESTi™-score. Data used for the radar diagram for gluteus superficialis can be found in Appendix N.

### 3.5.3 Descriptive data

No significant difference was found when clustering the dogs according to gender, weight, age, experience, athletic score, normal hurdle height in training and normal contact-method practiced on A-frame.

According to training level the dogs with 1 hour of training per week or more (n=6) had a significantly higher ( $p < 0.05$ ) E- S- and ESTi-score for triceps brachii both before (E=4.5, S=6.2, ESTi=5.9) and after (E=5.0, S=5.8, ESTi=6.1) 15 minutes of cool-down exercises compared to dogs with less than 1 hour of training per week (n=8) (before E=1.5, S=3.4 ESTi=3.8 and after E=1.6,

S=4.0 ESTi=4.2). Dogs with 1 hour of training per week or more recruited less muscle fibers in triceps brachii and were more effective and coordinated before and after warm-up exercises compared to dogs with less than 1 hour of training per week. No significant difference was found in gluteus superficialis. See appendix N for data.

According to height of the dogs, the dogs above 55 cm (n=7) had a significantly lower ( $p<0.05$ ) T- and ESTi-score for triceps brachii before the cool-down exercises (T=6.3, ESTi=4.0) compared to dogs at 55 cm or below (n=7) (T=7.4, ESTi=5.5). Dogs above 55 cm had a higher firing rate in triceps brachii before the cool-down exercises, but no difference was found after the cool-down exercises. No significant difference was found in gluteus superficialis. See appendix N for data.

## 4.0 Discussion

This is the first study of its kind to test AMG on very active dogs. Furthermore, it is the first study to investigate how warm-up and cool-down exercises, height of hurdles and A-frame effect the muscles of dogs participating in agility with the use of AMG.

### 4.1 Sources of error

Before discussing the results some sources of error shall be mentioned and it will be discussed how they may or may not affect the results. These errors shall be taken into account when discussing the results.

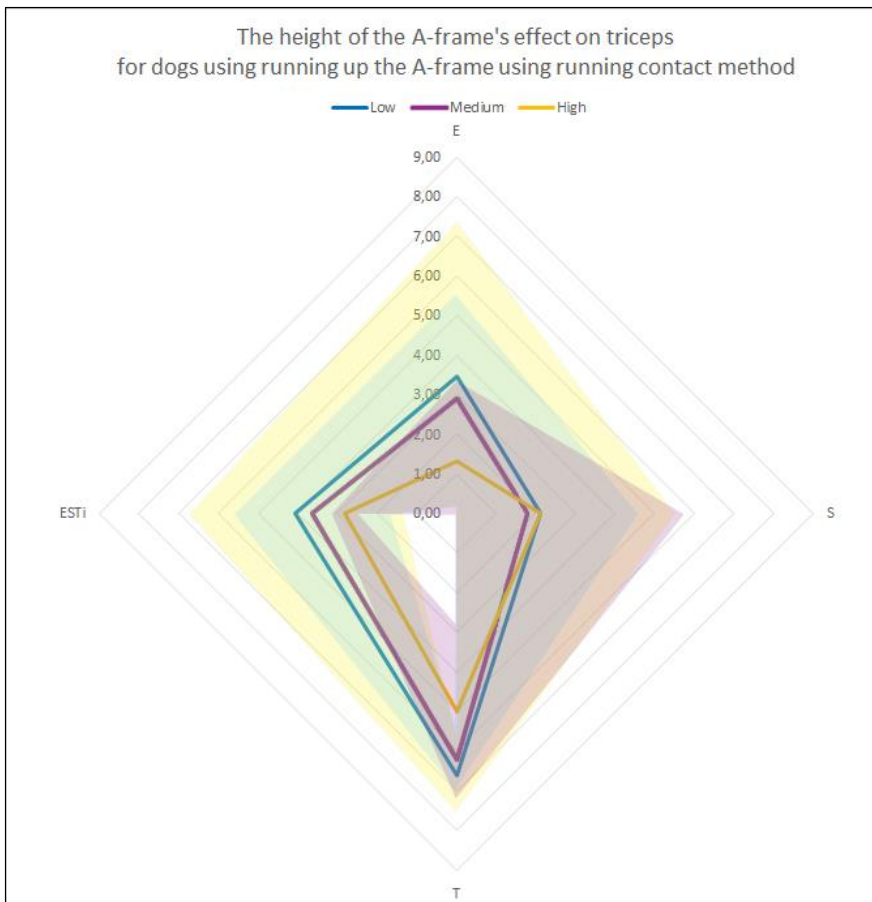
#### 4.1.1 Sample size and variance

14 dogs participated in the experiments however more dogs would have been preferred, especially when clustering the dogs in different groups for comparison. Three dogs practised running contacts resulting in a very little group to test for homogeneity. Additionally, the dogs were of different breeds, age, weight, heights etc. resulting in increased variance. Alcock *et al.* (2015) found that border collies might have a different jumping trajectory than other breeds. Using the same breed, at approximately the same age, size and training level could have minimized the variance thus making it easier to see a significant difference, but it would not represent the population of dogs performing agility. Thus, if the intention of only using border collies had succeeded it would merely have reflected how border collies jump.

The dogs and handlers performed the warm-up exercise with different intensity and duration. For example, some dogs galloped casually next to their handler while others galloped with high speed running for a ball resulting in a different intensity of work on the muscles being warmed up. Some dogs had never tried a cavaletti before, and it was clearly seen how they improved doing the task for each trial. To ensure a correct execution, some dogs did the cavaletti more than the prescribed three sessions. On the other hand, some dogs got tired of the slalom or the figure-eight movements between their owner's legs and simply just lied down resisting to do further repetitive boring exercises. This could result in a bigger variance in the result diminishing the possibility of finding a statistically significant effect of the warm-up exercises.

The absence of significant results for dogs used to practice the running contacts in table 10, 12, 12 and 13 may be due to the small sample size ( $n=3$ ) and the presence of a dog (Daphne) with odd data resulting in an increased variance. Daphne will be further discussed in section 4.7 *Daphne*. The increased variance is illustrated in picture 24 with standard deviation implemented explaining the lack of significant difference. Thus, the results from analysis of this group should be interpreted carefully for both muscles.





Picture 24: Radar diagram illustrating the average recorded ESTi™-scores and standard deviation on triceps brachii on dogs ( $n = 3$ ) running up the A-frame using running contact method. The dogs ran the A-frame in three heights 118 cm (low = light blue line), 148 cm (medium = blue line) and 167 cm (high = dark blue line) respectively. The axis from centre and out shows the ESTi™-score. The coloured areas mark the mean  $\pm$  standard deviation for each height in the respective colour. Notice: the colours are transparent so yellow and blue areas become green. Data used for the radar diagram for triceps brachii can be found in Appendix L.

#### 4.1.2 The analysis and statistics

Some results were sensitive to where you marked on the graph in the online CURO equine programme, and the ESTi™-scores could vary 1-2 scores when moving the cursor only few seconds according to the timeline. Likewise, the threshold was continuously changed according to the character of the graph, also resulting in different outcome of the ESTi™-scores. When a score of 0 was achieved on all the ESTi™-parameters the markings were changed without compromising the selection criteria. Although the selection criteria were not compromised, there could be a big variance depending on the markings and resulting in both higher and lower ESTi™-scores. As there would be no unidirectional influence the effect is unknown and a counterbalance could occur to some degree.

According to the statistics many tests have been made and the risk of false signals (Martinussen 2017) is high. It is impossible to identify which results is being a false signal, but contradicting results could be signs.

It should be noted that the statistic tests for the hurdles and A-frames are made on summary measures and it is not strictly statistically legitime to do so. It is however judged sufficient considering the level of the study and student (Martinussen 2017). Alternatively, summary measures could have been avoided by altering the design of the study by only allowing the dogs to run each obstacle once, but then the results would be more vulnerable to outliers.

#### 4.1.3 The AMG-equipment

The weight of the CURO (300g) is added to the anterior of the thoracic region of the dog and may affect the movement and use of the muscles. Besides investigating the effect of trotting up and downhill Carrier *et al.* (2008) investigated the effect of adding a mass of 0%, 8% and 12% of the body mass using fEMG on six mixed breeds when running. Besides a 3% reduction in the period of the swing phase when adding the 8% mass to the anterior trunk, they found no significant effect of adding mass to this area. The weight of the CURO ranged from 0.8% to 2.3% of the dogs' body mass. It is less likely that the weight of the CURO had an effect on the movement of the dog, but the sensory feeling of the CURO, sensors and Snøgg may have, although all dogs visually walked fine and seemed unaffected after only a few minutes.

Triceps brachii is capable of producing extremely large amount of force and is the strongest muscle in both the thoracic and pelvic region (Williams *et al.* 2008b). This is also found in this study with a high amplitude measured resulting in clip of amplitude as the maximal range of the 6dB sensors available at the time of this study was reached. In picture 27 the clip of the amplitude is visible already when jumping the low hurdles resulting in a S-score of 0. It is not possible to score below 0, meaning an increased S-score when jumping higher hurdles could not be measured by the AMG-equipment nor shown on the ESTi-score. When comparing the recordings in picture 27a and picture 27b an increased amplitude can be seen, most noticeable in the landing phase. Interestingly there seems to be a higher amplitude in the take-off when jumping low hurdles, when merely looking at this dog performing one jumping sequence. A visual analysis of all 14 dogs has not been made, but could have clarified differences that otherwise could not be measured by the AMG-equipment due to limitations of the maximum amplitude of the 6dB sensors. The same problem is evident when looking at triceps brachii when the dogs run the A-frame illustrated in picture 31. These limitations have affected the possibility of finding significant differences according to the recruitment of fibres and thus affect the overall improvement in triceps brachii. For this reason, the S and ESTi-score should be interpreted with caution, but when a significant difference is found it is despite of these limitations.

The same problem is not evident when measuring on gluteus superficialis as the sound from this muscle rarely exceed 6db, and ESTi<sup>TM</sup>-scores of 0 are not profound, but do occur. As with triceps

brachii a similar tendency is visible for a higher amplitude at take-off and lower for the landing phase when jumping higher compared to lower hurdles. Still both are assigned the S-score 4 and no significant difference was found. Perhaps with a range from 0-100 instead of 0-10 these differences could have been detected. Subsequent to completing the experiment, the CURO has been further developed and the score is now signed from 0.0-10.0 with a single number mantissa and 0dB sensors now available capable of measuring the full range of amplitude.

#### 4.1.4 Inexperience




Dogs were not experienced in jumping high hurdles (see hurdle height in table 6) and did not have a well-trained jumping trajectory on high hurdles resulting in knock-down of bars or refusal of jumping. Dogs that found it hard to jump high hurdles had a few tries and the handler often supported the dog further by running right next to it guiding the dog forward. This could affect the use of the muscle and e.g. result in a lower ESTi<sup>TM</sup>-score as a result of the unfamiliar use of the muscle. Instead higher E and ESTi-scores were found. It is possible that jumping higher hurdles compared to low makes the dogs more focussed on the task resulting in more coordinated use of the muscles and that they are simply careless and unfocussed when jumping low hurdles.

The dogs were neither used to running the lower heights of the A-frame and thus the effect of height found may only reflect the inexperience of the dogs rather than the actual effect of the height. This may explain why the dogs were more efficient when running down the high A-frame compared to the low A-frame. Furthermore, the lack of results may be due to the inexperience of the dogs running the low A-frame as it could be expected to find a higher ESTi<sup>TM</sup>-score with lower A-frame heights, but the inexperience of the dogs may counterbalance the effect resulting in no significant difference. To avoid this bias dogs with no experience of running the A-frame could have been chosen and trained in all heights or placed in three groups and each group trained in one height. They could also have been trained in practising both the running and 2o2o-contact methods to avoid similar bias in these results. This would have been a comprehensive work for this study, but the owners could have been asked to practise the different heights and contact method before entering the trials. This would probably result in fewer handlers volunteering in the trial.

The dogs were not used to practice the other contact method either and the inexperience may have resulted in a lower ESTi<sup>TM</sup>-score. For this reason, comparison of dogs used to practice the 2o2o-contact method (n=11) with dogs used to practice the running contacts (n=3) were made. Picture 25 illustrates this to help understand the comparison of these groups.

Continuing with the bias according to experience in the contact method, some were not sufficiently trained in the contact method used which resulted in some dogs not having a real

2o2o-stand before running the A-frame and the stand often became less marked when running the low A-frame. This could be the reason why no significant difference is found according to muscle recruitment and firing frequency as hypothesized. The 2o2o-contact method looked for some dogs more like running contacts. Meanwhile the protocol took this into account allowing helping tools such as a treat in a bowl or toys. Still some did not touch the contact area. Although the dogs performed the 2o2o and running contacts at very different levels this reflects the situation at a competition or training making the study more comparable to real-life situations.

Clusters	Score	2o2o-contact method		Running contact method		Test	p-value
		Mean	SD	Mean	SD		
Both contact methods n=14 	E	2,7	0,9	1,8	1,0	T-test	0,005
	S	0,7	1,4	0,7	1,7	Wilcoxon	0,779
	T	7,2	1,1	6,7	1,6	Wilcoxon	0,286
	ESTi	3,5	0,8	3,1	0,5	T-test	0,017
2o2o-contact method n=11 	E	2,8	1,0	1,9	0,7	T-test	0,008
	S	0,3	0,3	0,3	0,5	Wilcoxon	0,753
	T	7,6	0,7	7,2	1,1	Wilcoxon	0,351
	ESTi	3,6	0,6	3,1	0,5	T-test	0,012
Running contact method n=3 	E	2,4	0,8	1,3	2,0	T-test	0,388
	S	2,0	2,9	2,1	3,7	Wilcoxon	0,655
	T	5,8	1,6	5,0	2,2	T-test	0,732
	ESTi	3,4	1,5	2,8	0,3	Wilcoxon	0,593

Picture 25: Illustration of which groups are compared when comparing dogs used to practice the 2o2o-contact method (n=11) with dogs used to practice the running contacts (n=3). The data in the blue boxes are compared.

#### 4.1.5 Real-life situation

The dogs had walked a distance of approximately 160 meters from the car to the training area and moved a bit around before the dog was fully equipped. This could result in a higher initial ESTi<sup>TM</sup>-score making it less easy to see an improvement, although this would be the same at a normal training or competition situation, thus making the results more comparable with real-life situations.

Pfau *et al.* (2010) only looked at the effect when landing and other studies distinguish between the phases of jumping (Birch & Lesniak 2013; Birch *et al.* 2015; Cullen *et al.* 2017). In this thesis, the whole jumping trajectory over 5 hurdles is combined, thus the different phases of jumping cannot be distinguished. As an illustration to better understand the activation of the two muscles during the jumping trajectory the acoustic myographic signal recorded is compared with the coherent jumping sequence in picture 27 for triceps brachii and picture 28 for gluteus superficialis.

The choice of measuring when a dog performs five hurdles in a row was made to have a mean score for more than the performance of a few hurdles as done by Pfau *et al.* (2010), Birch &

Lesniak (2013) and Cullen *et al.* (2017). At the same time, it can be expected that the dog had a more normal jumping trajectory when performing several hurdles compared to the performance at competition and training. Meanwhile the muscle activity for galloping between hurdles was included in the measurements, and there is no division of the phases for each jumping trajectory as this would result in a comprehensive work. It could however have been interesting to compare with the findings of Cullen *et al.* (2017). The trials have been made to reflect the situations at competition and training in which the galloping phase between is a part of the exercise and may also be affected by hurdle height. Thus, although the galloping phase and jumping of the hurdles cannot be distinguished in the results, the result represent a sum-up of the effect of hurdle height both effecting all phases of jumping and galloping as seen in picture 29. The mean and range of frequency and amplitude measured is shown to guide future studies.

#### 4.1.6 Measurements on one side

The measurements were only made on the right side of the dogs, but the dogs may have a strong and weak side resulting in different ESTi<sup>TM</sup>-scores, but as it is the same muscle that is measured for each trial and a pairwise test is made this should not have a substantial influence.

The dogs land with the forelimb loaded asymmetrically depending on the leading and trailing limb (Meershoek *et al.* 2001; Pfau *et al.* 2010; Cullen *et al.* 2017), and triceps brachii are only measured on the right side. The same could be the case when running the A-frame. If the sensor is placed on the trailing limb a lower S or T-score may be measured as either more muscle fibres must be recruited or fired with a higher frequency to endure a higher vertical force compared to the leading limb (Pfau *et al.* 2010). No analysis of preferred leading limb or change of leading limb has been made in this study. The effect of the difference in loading of the forelimbs would only be a problem if e.g. the dogs persistently chose to use one forelimb when jumping low hurdles while using the other when jumping high hurdles, thus confounding the effect of hurdle height with preferred leading limb. This is not likely to be the case. It is more plausible that the dog either randomly shift between a leading limb or have a preferred leading limb, thus the effect of hurdle height can still be seen. When watching video recordings from the trials this appears to be the case.

#### 4.1.7 Fatigue

Although it is expected that the dogs should easily be able to perform each trial there is a risk of fatigue. While the composition of the trials for the hurdles to some extent takes this into account, the effect of the height of the A-frame is susceptible to be confounded by the effect of fatigue.

There were signs of fatigue when running up the high A-frame, which may have resulted in a lower average T-score of the three runs. When comparing the T-score for triceps brachii running up the A-frame for each height, the T-score is lowest for the high A-frame (picture 33). The average T-score for the first run was 7.7. When comparing the average T-score for the low A-frame with the average T-score for the first run on the high A-frame conducting a Wilcoxon signed ranks test for paired samples with two tails no significant difference was found ( $p=0.97$ ) (appendix N). The sign of fatigue when running up the high A-frame, does not seem to confound an effect of the height of the A-frame.

No effect was found on gluteus superficialis when running up the A-frame at different heights. Meanwhile signs of fatigue were found when running down the medium A-frame, as the T, S and ESTi-score were decreased from the first to the last run. This could have affected the chance of finding a significant difference between the heights. To investigate this, the mean T-, S- and ESTi-score for the low and high A-frame was compared with the mean T-, S- and ESTi-score for the first run on the medium A-frame conducting a Wilcoxon signed ranks test for paired samples with two tails. A significant decrease in muscle fibre recruitment was found in gluteus superficialis when running down the medium A-frame the first time compared to the mean of running the high A-frame three times ( $p=0.02$ ), but not the low A-frame ( $p=0.56$ ) (appendix N). The sign of fatigue has most likely confounded the effect of the height of the A-frame on gluteus superficialis when running down the medium A-frame compared to the high A-frame. This should be interpreted with caution as the data compared are both a summary measure and raw data.

It is possible that running first the high and secondly the medium A-frame tired the dogs and thus the fatigue was seen when doing the eccentric work running down the medium A-frame. The task of running the lowest A-frame might have been less challenging giving the muscles time to recover before again running the high A-frame practising the other contact method where no fatigue was seen. If this is the case the dogs should not practice the A-frame more than 3 times in a row, otherwise they may fatigue and be in increased risk of injury.

The dogs practising the 2o2o-contact method may merely be fatigued when practising the running contact method and the results may not be an effect of shifting method. This could, however, be rejected as there were no signs of fatigue when comparing the first with the last trial when changing the contact method. Additionally, an effect is seen when comparing the two groups of dogs used to practise each contact method for which fatigue does not have an effect.

To avoid the height of the A-frames being confounded by fatigue the order of the heights could be randomized and changed between each run, but as this is an extensive work to do both during the trials and in the following work of the data and statistical analysis this was not done. Using the



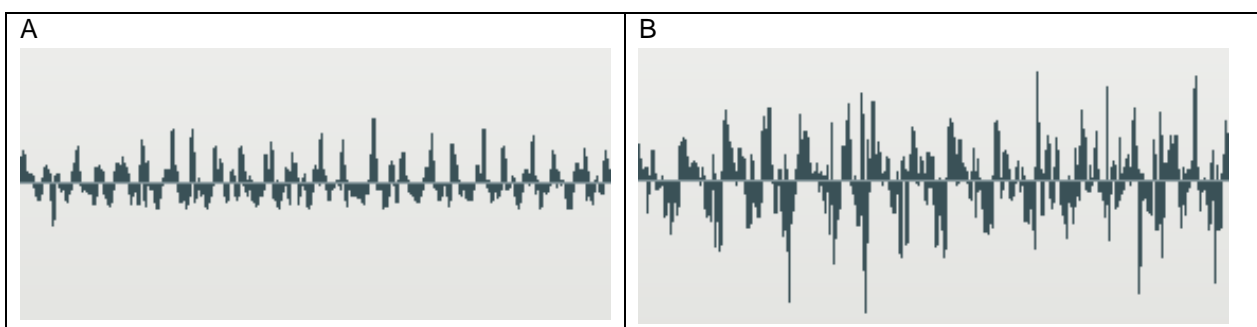
order in which the hurdles were taken enabled the possibility to test for fatigue more correctly, and would have been better to implement when testing the effect of the height of the A-frames, still it would have been labour requiring and statistically insufficient.

Knowing how the results may be affected by the sample size, variance, the analysis, statistics, the AMG-equipment, inexperience of the dogs, measurements done on only one side and signs of fatigue this can be taken into account when discussing the results. Future studies that manipulate these sources of error could shed further light on this matter.

## 4.2 The warm-up exercise

The dogs were generally better at using triceps when trotting after performing 10 and 15 minutes of warm-up exercises compared to when performing no warm-up exercises. Triceps was used significantly more efficient and coordinated after only 5 minutes and although there was no significant difference between 5-10 and 10-15 minutes for the E-score the significant differences between those and no warm-up exercise were increasing with increased time spent on warm-up exercises. This supports the theory of warm-up exercises improving neuromuscular coordination and increasing transmission speed of nerve impulses (Karvonen 1992). Several articles mention this effect of warm-up exercises and refer to Karvonen (1992) who refers to a popular scientific book written by Hill (1927) describing the function of muscles in the body. It has not been possible to get access to the original book, but newer studies using modern techniques to clarify the subject is recommended. Meanwhile this study shows that warm-up exercises improve coordination of the muscles, suggesting improved neuromuscular coordination and transmission of nerve impulses.

Warm-up exercises had no effect on gluteus superficialis. This might be due to the low workload when trotting as all scores were high from the beginning compared to triceps brachii resulting in little room for improvement. It is possible that an effect could have been observed with a higher workload and it could be interesting to make the same comparison when the dogs were galloping as gluteus superficialis showed a lower ESTi<sup>TM</sup>-score when galloping as illustrated in picture 26.



Picture 26: Example of an acoustic myography signal recorded from gluteus superficialis in a border collie trotting (A) and galloping (B) for 5.1 seconds.

Besides being able to measure the effect of warm-up exercises with the AMG-equipment, an effect of the warm-up exercises was visible. The dogs worked in a more focused and correct way for each repetition of the exercises, as seen with the cavaletti, although some dogs could be bored or tired after 15 minutes depending on the enthusiasm of the owner and dog and the monotony of the exercise.

Although some dogs got tired or bored after 15 minutes of warm-up exercises, it could be interesting to investigate the effect of an extended time of warm-up exercises. A tendency for the E- and ESTi-score to improve with increased duration of warm-up exercises can be seen and it is possible it increases further resulting in a significant difference. It could also be interesting to see when it levels out or results in fatigue. If this were to be done then an optimal duration of warm-up exercises could be found and recommendations made.

There was no sign of fatigue during the warm-up exercises, which would have been visualized with a decreased E-score and the warm-up exercises did not compromise the dogs' ability to perform the following trials. From this observation, it does not seem like the warm-up exercises reached the submaximal level of the dogs' capacity, which otherwise could have resulted in a lower energy turnover (Ferguson *et al.* 2002) and reduced their ability to perform (Gregson *et al.* 2002).

#### 4.2.1 Descriptive data

Besides the training level of the dogs no other characteristics of the dogs showed a significant difference according to warm-up exercises. The training level of the dogs will be further discussed in section 4.6 *Training level*.

#### 4.2.2 Risk of injury

The findings of an improved efficiency and coordination of the muscles, suggest that neuromuscular coordination is improved, and it could be hypothesized that a more efficient use of the muscles results in fewer agility-related injuries. Thus, the performance of warm-up exercises may reduce agility-related injuries. Meanwhile this have not yet been proved (Cullen *et al.* 2013a; Zachary *et al.* 2014; Huneycutt & Davis 2015), although it is suggested and theoretically explained that they do so (Strickler *et al.* 1990; Malone *et al.* 1996; Steiss 2002). Further investigation is needed to prove the effect of warm-up exercises on the risk of agility-related injuries, preferably under more controlled experiments than a retrospective study. Such an experiment is hard to execute as agility-related injuries only occur 1.7 injuries per 10000 hours (Zachary *et al.* 2014).

### 4.2.3 Conclusion

The hypothesis of triceps brachii and gluteus superficialis showing a higher ESTI™-score as a result of the performance of warm-up exercises is rejected for gluteus superficialis, but cannot be rejected for triceps brachii.

From the results of this thesis it is recommended to perform 15 minutes of warm-up exercises as it improves the overall use of triceps brachii, but the first 5 minutes are the most crucial in relation to the coordination of the muscle. This may result in fewer agility-related injuries, but is not proved to do so. This part of the study was merely an addition to the main study, but the possibilities of the findings, which by the knowledge of the author is the first of its kind for decades, arises numerous possibilities for further studies in the area with the use of AMG.

## 4.3 Hurdles

No similar studies have been made to study the effect of hurdle height on the muscles of dogs, but Pfau *et al.* (2010) found a higher workload on the muscles as well as a steeper landing angle when jumping high hurdles compared to long jump. From this a higher recruitment of fibres or higher firing rate could be expected, but this is not the case in triceps. Although not significant there was a general decrease in firing rate and muscle fibres activated as well as an improved coordination in triceps when jumping high hurdles compared to low, and the difference became significant when a combined ESTi-score was calculated. In general, the use of the triceps improved when the dogs jumped higher hurdles. Gregersen and Carrier (2004) found that the extensor muscle for elbow joints may store and recover elastic energy in dogs jumping with maximum performance. It is possible that the improved use of triceps brachii in this study is a result of the muscle, as an elbow extensor, using elastic energy when jumping high hurdles. Meanwhile, the same may not be seen with the low hurdles as a decreased flexion is seen in the radio-humeral and scapula-humeral joints at take off with lower hurdle height (Birch & Lesniak 2013), and may not store as much elastic energy. It is also found that storage of elastic energy in the distal forelimb of horses requires high tendon forces (Harrison *et al.* 2010). This may explain why an increased ESTi-score is seen for high hurdles compared to low hurdles as a higher load is put on the tendons enabling storage of elastic energy. When landing from high hurdles the stance time is decreased (Pfau *et al.* 2010), which means a decreased time for the energy to be absorbed by the front limbs. This could potentially be stored as elastic energy, but no effect on joint angles when landing according to hurdle height has been found (Birch & Lesniak 2013). Meanwhile a higher accelerative force with increased hurdle height have been found (Pfau *et al.* 2010). It is possible that the vertical force when landing is absorbed as elastic energy stored in the tendons, muscles and collagen of the front limb and then converted to forward kinetic energy in the following stride (Pfau *et al.* 2010).

The results from gluteus superficialis were in agreement with the expectation from Pfau *et al.* (2010) as the dogs generally recruited more fibres and had a higher firing rate in gluteus superficialis when jumping higher hurdles although not at a significant level. At the same time, they became significantly more effective and coordinated at using gluteus superficialis when jumping high hurdles compared to low hurdles, with the result of a stable ESTi-score. Opposite triceps brachii this does not seem to be a result of the storage and recovery of elastic energy as the hip joint only extends during take-off (Gregersen & Carrier 2004). Meanwhile, it is possible that the hind limbs like the front limbs use the elastic energy during landing, although 60% of the energy when landing is absorbed by the front limbs, and the front limbs take most of the decelerative force absorbing more energy (Pfau *et al.* 2010).

#### 4.3.1 Descriptive data

When analysing the data and dividing dogs into groups according to their descriptive data no effect was found of height or age which is in accordance with previous findings (Levy *et al.* 2010; Cullen *et al.* 2013). Zachary *et al.* (2014) did not take weight into account when analysing risk of injury as it was highly correlated with height, which Birch *et al.* (2016) also found to be the case. Cullen *et al.* (2017) did not consider weight, height or age at all when analysing the effect of jumping hurdles although comparison with their result could have been interesting as the study is quite similar. Meanwhile Birch & Lesniak (2013) expressed concern for dogs jumping hurdles which are substantially greater than the height of their withers due to a significant increase in extension of the sacroiliac region and a flexion in the scapula-humeral joint resulting in excess strain on their soft tissues. Birch *et al.* (2016) found a decreased length of jumping trajectory when dogs jumped >126% of their height suggesting signs that the dogs were approaching their limits. No effect of the height of the dogs was found when dividing the dogs into two groups separated at 55 cm. This could be due to the high height the dogs are divided at. In this study, the dogs jumped 102-141% of their height when jumping the high hurdle and the three lowest dogs (>51.6kg) jumped  $\geq$ 126% of their height.

Although no effect of the height of the dogs was found, dogs weighing 21 kg or less recruited more muscle fibres in gluteus superficialis when jumping medium and high hurdles compared to heavier dogs. No correlation has been analysed between weight and height in this study, thus this cannot be taken into account. Without taking the correlation with height into account it is preferred to have a dog with a weight above 21 kg when jumping hurdles above 45 cm as they do not need to recruit additional muscle fibres. The recruitment of fibres in dogs weighing 21 kg or less was close to its maximum potential as the S-score became closer to 0 (appendix K), thus the dogs are in increased risk of injury as maximum capacity of their muscles was reached. Due to the amplitude being clipped the maximum capacity for muscle recruitment may not be assessed. This does not

mean that smaller dogs should be overweight to reach the preferred weight and minimize fibre recruitment as all dogs in this experiment had a condition score of 4-5. It could be considered to implement the weight when distributing the dogs in their classes and not only their height at the withers (Birch *et al.* 2016).

Although some dogs were not used to jump high hurdles, there was no difference when comparing dogs used to jump hurdles of 55 cm or more with dogs used to jump lower hurdles. The inexperience according to hurdle height did not affect the results. Still, an effect was found for the training level of the dogs and this will be further discussed in section 4.6 *Training level*.

#### 4.3.2 Risk of injury

The force needed when jumping high hurdles is higher (Pfau *et al.* 2010), but it is met with an increased coordination of gluteus superficialis and overall improved use of triceps. Thus, the risk of injury when jumping high hurdles cannot convincingly be concluded to be higher compared to low hurdles. Instead the results indicate the opposite could be the case as the dogs might get careless. When dogs are 7 years of age or above the handler has the possibility to enrol a senior class with hurdles being maximum 45 cm high (Dansk Kennel Klub 2014). In the light of current results this cannot be recommended to do before strictly necessary as the dogs get less coordinated, and instead of easing the run for the dogs they may be more prone to injuries. No analysis of risk of injuries in senior dogs has been made to confirm this theory.

More weight is loaded on the forelimb and a shift of weight distribution towards the forelimbs (60%/40%) happens with increasing height of a hurdle (Pfau *et al.* 2010). The effect of this is not seen in the ESTi<sup>TM</sup>-scores for the two muscles, although it could be expected to observe a decreased ESTi<sup>TM</sup>-score for triceps brachii when the muscle has to deal with the extra weight in the landing phase. When comparing the recordings in picture 27a and picture 27b an increased amplitude in the landing phase can be observed, but may not be presented in the ESTi<sup>TM</sup>-score for triceps brachii due to amplitude > 6 dB being clipped. A high risk of injuries in the front limbs and especially the shoulder is found (Levy *et al.* 2013; Zachary *et al.* 2014), and the site of injury may be explained by this increased load on the forelimbs although it is not seen in the ESTi<sup>TM</sup>-scores.

The dogs may store and recover elastic energy when jumping high hurdles to deal with the increased force needed to take the jump and hence avoid injuries. Although the use of elastic energy is more energy efficient, it is also thought to increase the likelihood of injuries, as the distal forelimb in horses experience the highest loads (Harrison *et al.* 2010). In dogs, the injuries were significantly higher in shoulder, stifle joint, carpal joint and antebrachium when jumping hurdles (Cullen *et al.* 2013b) most of which is situated in the front limbs. The ability to store energy in the tendons make the force control easier, but at the expense of position control (Alexander 2002).

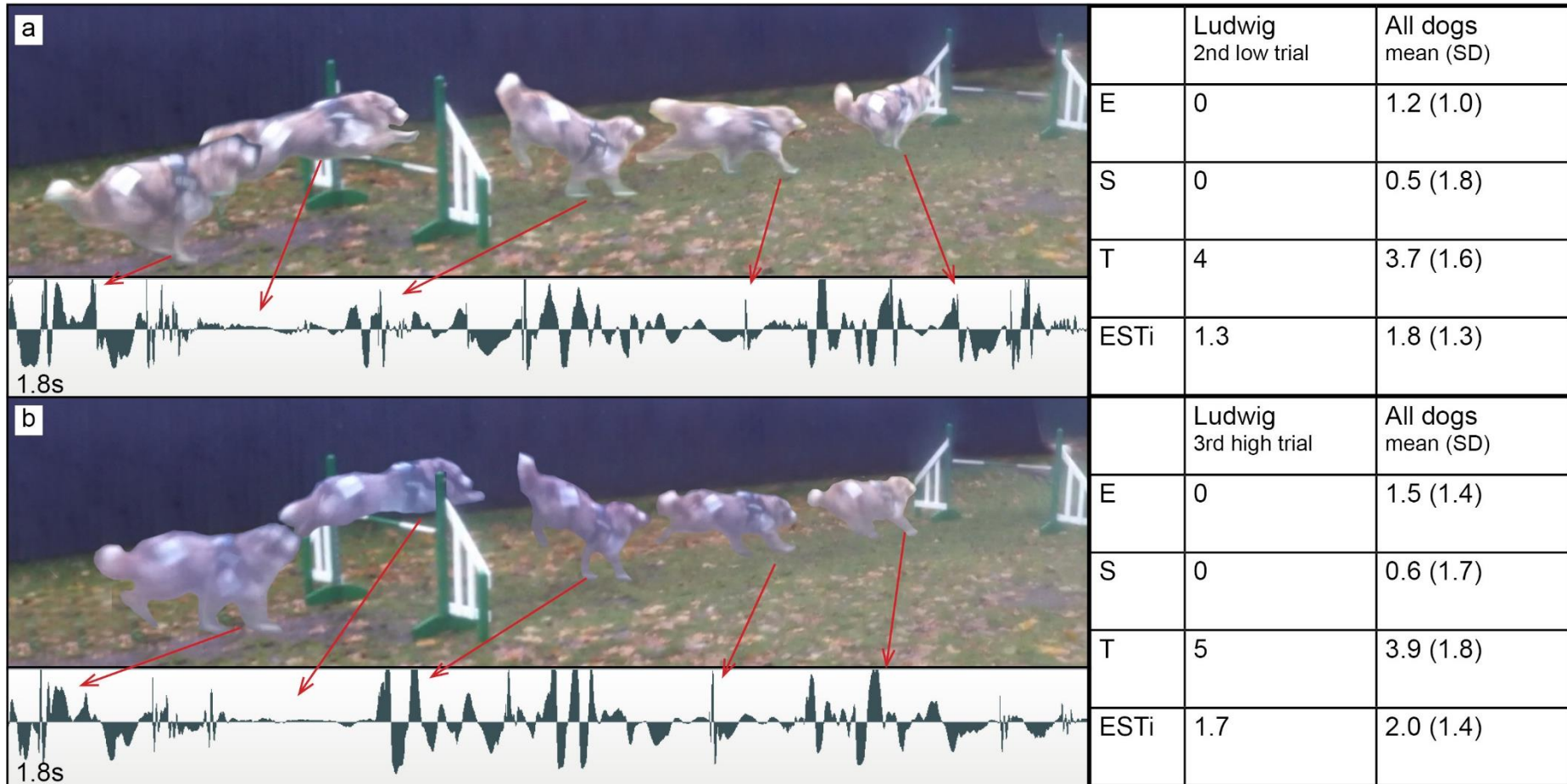
Although it is more energy efficient to use store and recover elastic energy it comes with expenses, and it may not be sufficient to avoid injuries during high forces.

The repetitive nature of jumping hurdles may contribute to a higher risk of injury (Pfau *et al.* 2009; Levy *et al.* 2013), but no sign of fatigue was shown in the trials nor signs of the muscles not being able to handle the different heights of the hurdles. From these results, there are no signs of a higher risk when jumping hurdles and no reason to have fewer hurdles on a course. The trials consisted of 5 hurdles while at competition the average is 13 hurdles plus the other obstacles being performed in a course (Ehrenreich 2017). Rodrigues *et al.* (2014 cited in Birch *et al.* 2016) found a decrease in jumping efficiency for horses when the number of hurdles increased, but this may not be similar for dogs which do not have a rider upon their back. It is possible that an effect of altered hurdle height can be seen when running an entire course as the effect of the repetitions as well as the possibility of fatigue could be measured. This could be interesting to investigate although tight turns and running through tunnels and weaving poles and other obstacles would be irresponsible when wearing this AMG-equipment as entangling is too high a risk. At the same time, the course of the hurdles was straight forward, and on an agility course the hurdles may be taken with tight turns thus loading the limbs and muscles differently. The dog may be asked to jump at unfamiliar angles or jumping close the hurdles thus more vertical force is loaded on the limbs, possibly asymmetrically. This would be relevant to investigate in relation to an increased risk of injuries (Birch & Lesniak 2013), although it may not be possible with the use of AMG due to risk of entangling of wires.

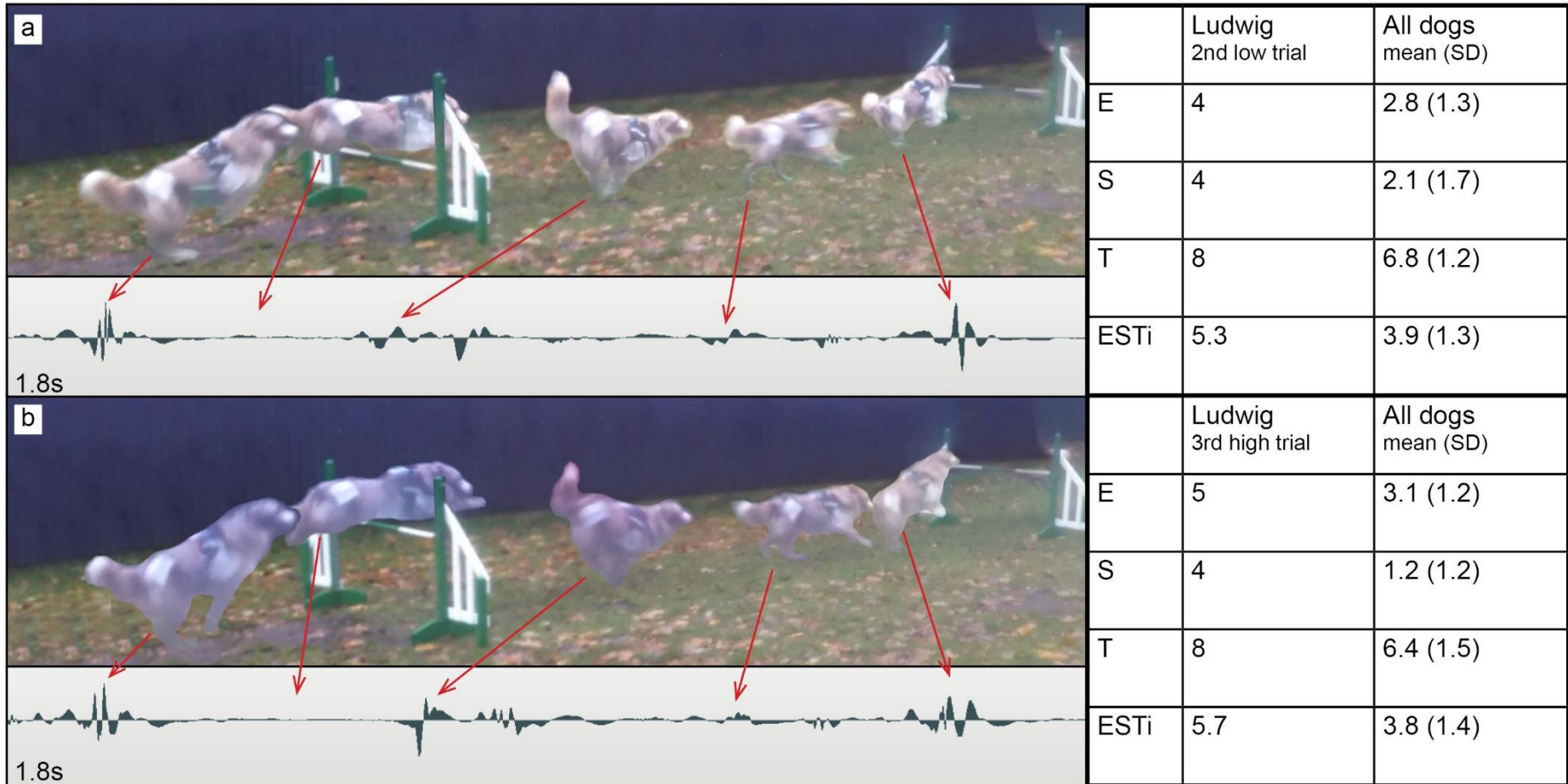
#### 4.3.3 Conclusion

The hypothesis of triceps brachii and gluteus superficialis showing a lower level of muscle engagement when jumping lower compared to higher hurdles is rejected for both muscles. The dogs used their muscles more coordinated and effective when jumping hurdles with a height of 65 cm compared to 45 cm. From this it cannot be recommended to lower the heights of the hurdles, but it may be recommended to classify dogs according to height as well as weight. When buying a future agility dog, it could be a good idea to buy a breed that ensures a final weight above 21 kg.

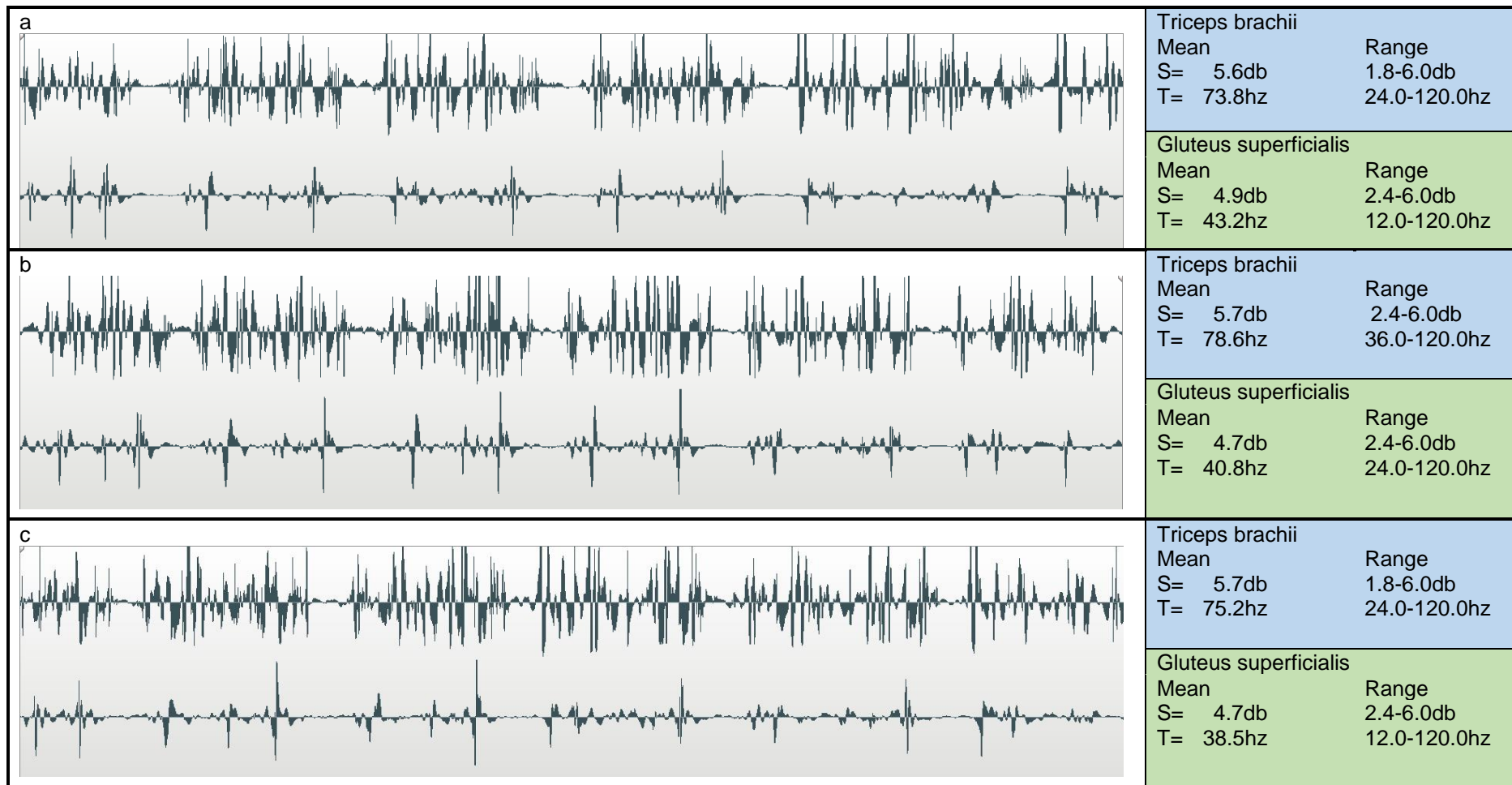




Picture 27: Illustration of a jumping sequence of a dog compared with the recorded acoustic myographic signal of triceps brachii. Picture a illustrates the jumping sequence and recordings when an Australian shepherd (Ludwig) jumps a 45cm high hurdle in a sequence of 5 hurdles for the second time. Picture b illustrates the jumping sequence and recordings when Ludwig jumps a 65cm high hurdle in a sequence of 5 hurdles for the third time. The ESTi™-scores are listed to the right for both Ludwig in the current jumping sequence and for the average of 15 dogs jumping the sequence three times together with standard deviation (SD). The pictures are taken with a Iphone4 or 5 camera explaining the poor quality and the pictures should be interpreted carefully.



Picture 28: Illustration of a jumping sequence of a dog compared with the recorded acoustic myographic signal of gluteus superficialis. Picture a illustrates the jumping sequence and recordings when an Australian shepherd (Ludwig) jumps a 45cm high hurdle in a sequence of 5 hurdles for the second time. Picture b illustrates the jumping sequence and recordings when Ludwig jumps a 65cm high hurdle in a sequence of 5 hurdles for the third time. The ESTi™-scores are listed to the right for both Ludwig in the current jumping sequence and for the average of 15 dogs jumping the sequence three times together with standard deviation (SD). The pictures are taken with a Iphone4 or 5 camera explaining the poor quality and the pictures should be interpreted carefully.



Picture 29: Example of an acoustic myography signal recorded in an interval of 6.9 seconds from triceps brachii (upper graph) and gluteus superficialis (lower graph) in a border collie (Ludwig) jumping 65cm (a) 55cm (b) and 45cm (c) high hurdles. For each height and muscle the range and average of S and T-scores have been translated to amplitude and frequency respectively for 14 large dogs jumping 5 hurdles.

## 4.4 The A-frame

### 4.4.1 The effect of the height

#### 4.4.1.1 *Triceps brachii*

Cullen *et al.* (2017) did not find a significant difference in triceps brachii when comparing a high (1.75 meter) with a low (1.67 meter) A-frame, and the findings in this thesis were similar. The only effect found in triceps when changing the height of the A-frame was an improved coordination when running down the high A-frame compared to low. The effect was found when the usual contact method was not taken into account, which Cullen *et al.* (2017) neither did. Similar results were found when comparing effect of hurdle height on triceps as discussed in the previous section for which a general improved use of the muscle with increased hurdle height was measured. As suggested earlier it is possible that the dogs simply get careless when not being challenged sufficiently in their performance. Meanwhile, it could also be due to the unfamiliar height. The running trajectory of the A-frame changes when the A-frame is positioned at a low height compared to the high and medium height as illustrated in picture 33. The dogs are accustomed to run the high A-frame and they are not trained to run a low A-frame, and hence the coordination of their muscles may not be as effective resulting in a lower E-score. Following an BMX-rider for 10 months showed a rapid improvement in the brain figuring out how many fibres to fire and at what frequency (Bartels *et al.* 2017). Meanwhile, the coordination of the muscles improved less rapidly, but continuously during the 10-month period. Thus, it takes several months for muscles to get more coordinated, and simply running the A-frame a few times would not improve the coordination, while the high A-frame is well-trained and the coordination effective. Although training on the low A-frame could be considered a method to minimize the impact on the musculature on the dogs during training allowing more runs this is not the case. The dogs should be trained if altering the height of the A-frame starting with low level intensity, not training them as they usually would with the high A-frame.

No significant differences have been found when looking at dogs used to practise running contacts. Still it is interesting that a generally higher ESTi<sup>TM</sup>-score can be observed when running down the medium A-frame suggesting this to be the most optimal size of the A-frame when using running contacts. These results are not caused by the dogs used to run a medium A-frame. Further studies with a bigger sampling size should be made before making a conclusion on this.

When looking at picture 31 it is clearly visible that additional muscle fibres are recruited for a shorter time when running up the A-frame compared to running down. This is similar with the findings of Carrier *et al.* (2008) who found an increased activity in forelimb muscles when trotting 14° uphill and decreased activity in some when trotting 14° downhill. When looking at the ESTi<sup>TM</sup>-

scores all scores are increased when running down meaning triceps brachii are more coordinated, less muscle fibres are recruited and at a lower firing rate. The ESTi™-score was increased no matter the height of the A-frame or the contact method used as illustrated in picture 33, although in Jamie's case it is lower when running the low A-frame. Similar observations can be made for gluteus superficialis in picture 32 and 34. Although no statistical test has been made for this observation Cullen *et al.* (2017) found similar results, as triceps had the highest peak activation when running up the A-frame, while the lowest peak activation when running down the A-frame. Every participating dog used two steps running up the A-frame thus all power needed to get up is gathered in only two steps. In contrast, it is divided in several smaller steps and movements when running down the A-frame especially when practising the 2o2o-contact method. Thus, the energy needed to run down the A-frame is distributed over several steps partly the reason of the increased ESTi™-score. The change of ESTi™-scores could also be explained by the change from the muscles doing a concentric exercise and propulsive work running uphill to an eccentric exercise and making a breaking force running downhill (Carrier *et al.* 2008; Lauer *et al.* 2009).

#### 4.4.1.2 *Gluteus superficialis*

It could have been expected that the practice of contact method merely affected the run down the A-frame and not up. This is not the case in this study. The dogs practicing the 2o2o-contact method recruited more muscle fibres in gluteus superficialis when running up the high A-frame compared to the low A-frame. It is possible that the dogs lower their speed up the A-frame to prepare for the breaking force down the A-frame and thus triceps brachii may be used for more propulsive work resulting in more muscle fibres recruited. The dog does not need to prepare when going uphill with a lower A-frame, as the need for breaking work down the low A-frame eases, resulting in fewer muscle fibres recruited. Otherwise no effect of height of the A-frame was found which is similar with the findings of Lauer *et al.* (2009) although they compared a 5%, 0% and -5% inclination. In this study, the inclinations were: high: 81%, medium: 65% and low: 49%. Robert *et al.* (2000) found an increased muscle activity in gluteus medius in horses running a 6% inclination. Although it was suggested that gluteus superficialis may show the same result, this was not the case and it was measured on another species.

While the running trajectory is similar comparing the medium and high A-frame, the whole running trajectory of the A-frame changes when the A-frame is positioned at a low height as illustrated in picture 34. This could have affected the muscles, but did not result in a significant change of how the gluteus superficialis was used.



#### 4.4.1.3 Descriptive data

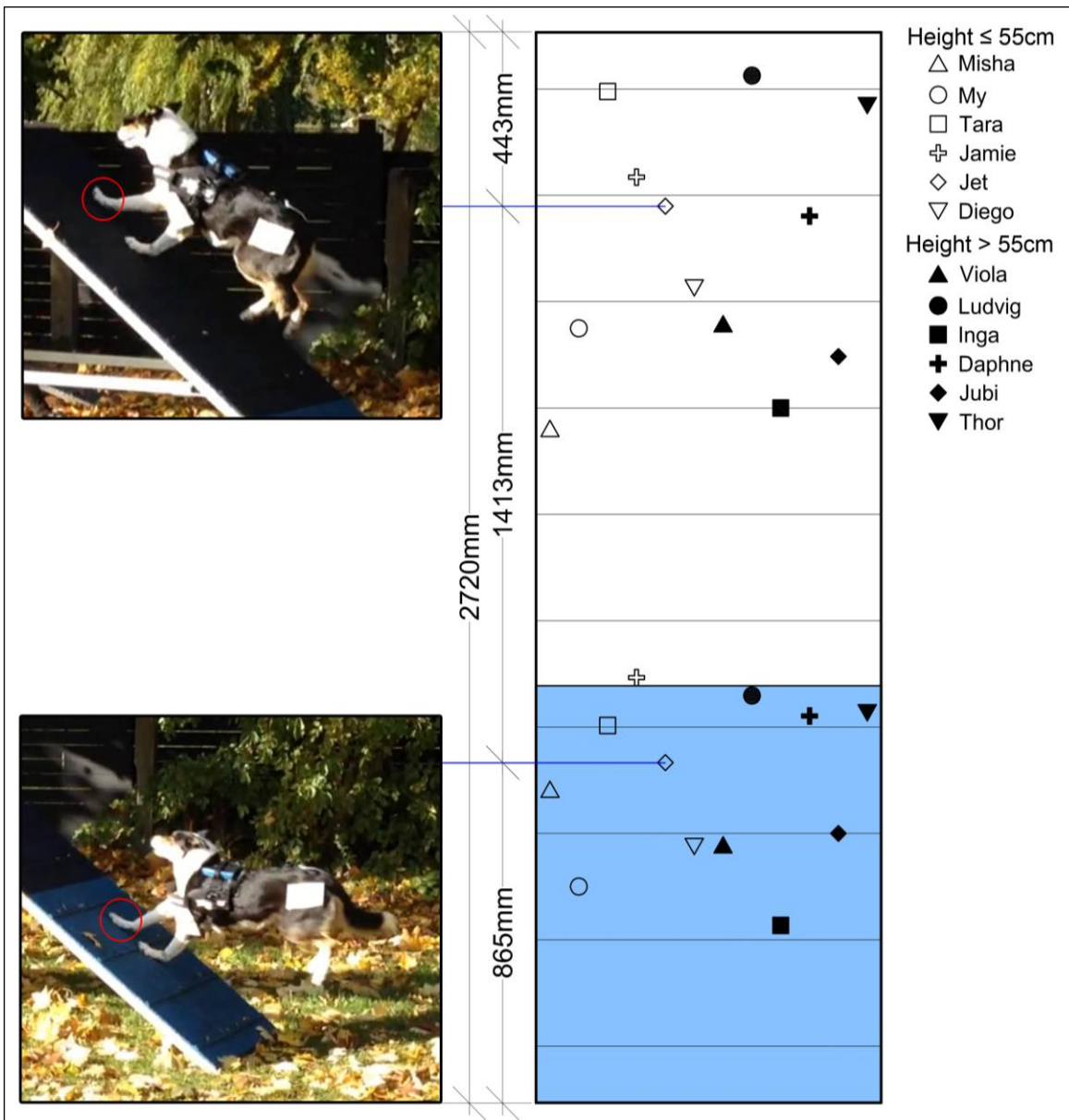
Taller dogs (> 55 cm at the withers) seem to be more challenged when running up the high and medium A-frame and this is met with an increased firing rate. It is possible that taller dogs have altered joint angulations when running the A-frame, explaining the need for a higher firing rate. Analysis of kinetics and joint angulations when running the A-frame could clarify this point. An altered jumping technique was found with increased percentage height (Birch *et al.* 2016) and the same could be the case when dogs in different heights run a high A-frame. It is not possible to investigate joint angulation etc. from the data and recordings in this thesis, but the position of the paw on the ramp when running up the A-frame is visible on the video recordings for 12 dogs. Generally, the length and height of the ramp when running up the A-frame is proportionally lower for the higher dogs compared to the lowest dogs. It could be hypothesised that higher dogs position their paws in another distance as higher dogs normally have longer strides (Fisher & Lilje 2014). The stride-length and position of the right front limb on the ramp was measured using video recordings as shown in picture 30 to investigate if the highest dogs had an altered running trajectory up the A-frame. The result is listed in table 14 and no significant differences were found when comparing the two groups of heights. Thus, it can be rejected that dogs above 55cm have longer stride-length when running up the high A-frame, and they do not encounter or leave the A-frame in an altered position compared to the lower dogs. On the contrary, the results suggest that the higher dogs may shorten their stride-length relative to the lower dogs. This implies a lower range of motion and joint angulation in dogs above 55cm when running up the high A-frame. This may result in an increased force needed during the lower range of motion which the brain regulates by increasing the rate with which the muscles are fired.

Training level had an effect according to the height of the A-frame and this will be discussed in 4.6 *Training level*.

Table 14: Twelve dogs running up a high A-frame (170cm) are divided into two groups according to their height at the withers, height  $\leq$  55cm (n=6) and height > 55cm (n=6). Distance between the two touchdowns of the right front paw and distance from bottom of the ramp to the first touchdown and from top of the ramp to the last touchdown is measured and mean and standard deviation calculated. The two groups are compared using a Wilcoxon Rank-Sum tests with p-values listed in the table. The data can be found in appendix N.

	Dogs height $\leq$ 55cm		Dogs height > 55cm		p-value
	Mean	(SD)	Mean	(SD)	
Distance:					
- between touchdowns	134cm	(23cm)	137cm	(15cm)	0.749
- from bottom of the ramp to the first touchdown	82cm	(19cm)	80cm	(24cm)	1.000
- from top of the ramp to the last touchdown	56cm	(31cm)	55cm	(35cm)	1.000





Picture 30: Position of the right front paw of 12 dogs trained in agility when running up a high (170cm) A-frame. Unfilled markings are dogs with a height at the withers between 43 and 55 cm while filled markings are dogs above 55 cm. The blue area is the contact area and is positioned in the bottom 106 cm of the ramp. Two pictures show the position of the right front paw on a border collie (Jet), and distances between the two points as well as distance to the top and bottom of the ramp are shown.

#### 4.4.1.4 Conclusion

The hypothesis of triceps brachii and gluteus superficialis showing a lower level of muscle engagement when running an A-frame with a lower height and slope compared to a higher and steeper A-frame is rejected for triceps brachii, but cannot be rejected for gluteus superficialis. The dogs used triceps brachii in a more coordinated and effective manner when running down the A-frame with a height of 170 cm compared to 120 cm. Meanwhile the dogs practicing the 2o2o-contact method recruited more muscle fibres in gluteus superficialis when running up the high A-frame compared to the low A-frame. Based on these results it cannot undisputed be recommended to lower the A-frame.

## 4.4.2 The effect of contact method

### 4.4.2.1 *Triceps brachii*

Triceps brachii is used more efficiently and coordinated when practicing the 2o2o-contact method running both up and down the A-frame. However, it may not be the case for dogs used to practice running contact method as no significant differences were found when they shifted to the 2o2o-contact method (see table 10 and 11). This may be due to the small sampling size as the differences were similar to that of dogs practicing 2o2o-contact method. Meanwhile, when comparing dogs used to practice the 2o2o-contact method (n=11) with dogs used to practice the running contacts (n=3) (picture 25) similar results are found when running up the A-frame. Thus, practicing the 2o2o-contact method improved the use of triceps brachii when running up the A-frame and using this method may reduce the risk of injury. Although a difference would be expected when running down the A-frame no differences were found when comparing dogs used to practice the 2o2o-contact method with dogs used to practice the running contacts. Thus, it may only be the shift from practicing a well-trained method to an unfamiliar method that effects the dogs when running down the A-frame instead of the actual contact method used. Meanwhile the fact that a significant difference is seen when clustering all the dogs together contradict this concern.

### 4.4.2.2 *Gluteus superficialis*

No significant difference was found in gluteus superficialis when running up the A-frame, no matter the contact method practiced or how they were compared. Running down the A-frame the dogs used gluteus superficialis more efficiently when using the 2o2o-contact method when the accustomed contact method is not considered. Meanwhile, when considering the usual contact method used, dogs used to practice the 2o2o-contact method were generally less coordinated when shifting to the running contact method.

Although it was hypothesized that higher ESTi<sup>TM</sup>-scores would be seen when practising running contacts compared to 2o2o-contact method, due to a smoother running-technique over the A-frame without a full stop at the end, this was not the case. It is possible that dogs practising the 2o2o-contact method have to think more about their use of the muscles and coordination to prepare for the full stop, while dogs who are simply running over the A-frame do not have to be that coordinated. This could explain the difference in the E-score. Meanwhile no increased fibre recruitment or higher firing rate is necessary.

### 4.4.2.3 *Conclusion*

The hypothesis of triceps brachii and gluteus superficialis showing a lower level of muscle engagement when using a running contact method compared to the 2o-2o-contact method when

running the A-frame is rejected. Practicing the 2o2o-contact method improved the efficiency and overall coordination in triceps brachii when running up the A-frame, and similar findings were seen when running down for both muscles. Dogs used to practice the 2o2o-contact method should not change to practicing running contact without prior training. Meanwhile there is no reason not to change between the practice of contact method when a dog is trained in the running contact method. How these results may affect the risk of agility related injuries will be discussed in the following.

#### 4.4.3 Risk of injury

A lower A-frame may increase the risk of injury as the dogs might get careless, otherwise the adjusted height of the A-frame should be trained for the muscle coordination to be adjusted.

A higher muscle fibre recruitment is visible in dogs running up the A-frame compared to running down. It is possible that triceps brachii is close to reaching maximum capacity and the increased muscle fibre recruitment shows how much force is directed on the front limbs running up the A-frame. The higher than expected number of injuries in the shoulder and phalanges (Cullen *et al.* 2013) may partly be caused by running up the A-frame.

Another explanation of the high number of injuries in connection with running the A-frame could be the eccentric work executed when running downhill (Lindstedt *et al.* 2001; Lauer *et al.* 2009). When looking at picture 31 and 32 the eccentric work when running downhill can be seen with the force needed distributed over several steps compared to the concentric work shown using two steps when running uphill. When doing eccentric work a lower number of type II fibres are recruited and the force is taken up by the type I fibres which are unable to cope with the considerable force produced by the type II fibres and as a result are more prone to injuries (Friden & Lieber 1992; Harrison 2017b). The long head and lateral head of triceps brachii has a relatively high percentage of type II fibres as well as gluteus superficialis, while the opposite is the case for the accessory head and medial head of triceps brachii. Type II fibres contract fast and are easily fatigued, but when running down the A-frame the stance phase is prolonged compared to the swing phase (Cullen *et al.* 2017) and thus a prolonged need for muscle fibres to be active occur. Instead type I muscle fibres must take over not being the optimal type for eccentric contractions and less abundant in gluteus superficialis and the two biggest heads in triceps brachii. If the muscles cannot meet the needs during eccentric contractions injuries may occur, and the eccentric work weakens when the muscle has previously been damaged (Jonhagen *et al.* 1998; Brockett 2004).

Dogs used to practise the 2o2o-contact method may be at increased risk of injury when shifting methods as they impaired their use of triceps brachii both running up and down the A-frame.

Their muscles are not familiar with the pattern, and a shift in pattern when running down the A-frame while doing eccentric work often causes injuries, but with proper training this can be avoided (Lindstedt *et al.* 2001). Although no reason is found not to shift between the practice of the contact method when trained in practicing the running contact method, this should be done with caution, as a shift from the normal pattern occurs.

The practise of the 2o2o-contact method may reduce the risk of injuries in the hind and front limbs as a significantly improved use of the two muscles is seen. Cullen *et al.* (2013) did not distinguish between the practiced contacts method when investigating the risk of injuries, type and location in relation to the A-frame. Generally, no comparison of the two contact methods has scientifically been investigated before, making these results the first of their kind. Based on the present results, it is possible that the muscles are better at handling the slower decelerative work down the A-frame with the full stop (Carrier *et al.* 2008; Cullen *et al.* 2017), than the work of converting potential energy to forwards kinetic energy.

The A-frame is usually only used once in a course at a competition, but may be practised several times when training. Repeated movement with eccentric contraction may accumulate macro damages to muscle fibres that predispose to injuries (Chumanov *et al.* 2007; Schache *et al.* 2009; Schache *et al.* 2010). This could result in injuries both when running the current obstacle, but as macro damages may take place an increased risk of injury is carried on to the following obstacles on the course. Injuries occurring on e.g. hurdles may be partly due to damage on the muscle from running the A-frame earlier on the course. The opposite could be the case as well. In this study, the dogs ran the A-frame a total of 12 times, half of them at the highest position. Meanwhile no apparent injuries were observed and the most apparent sign of fatigue was in gluteus superficialis when running down the medium A-frame. As mentioned earlier it is possible that the dogs should not practice the A-frame more than 3 times in a row, otherwise they may fatigue and be in increased risk of injury.

Some effect was found in the two muscles, but further studies with a higher sampling size and randomization of the trials are recommended together with a more even distribution of dogs using the two contact methods as well as trained at each height and contact method. To aid future studies the range and average of S and T-scores have been translated to amplitude and frequency respectively as seen in table 15.

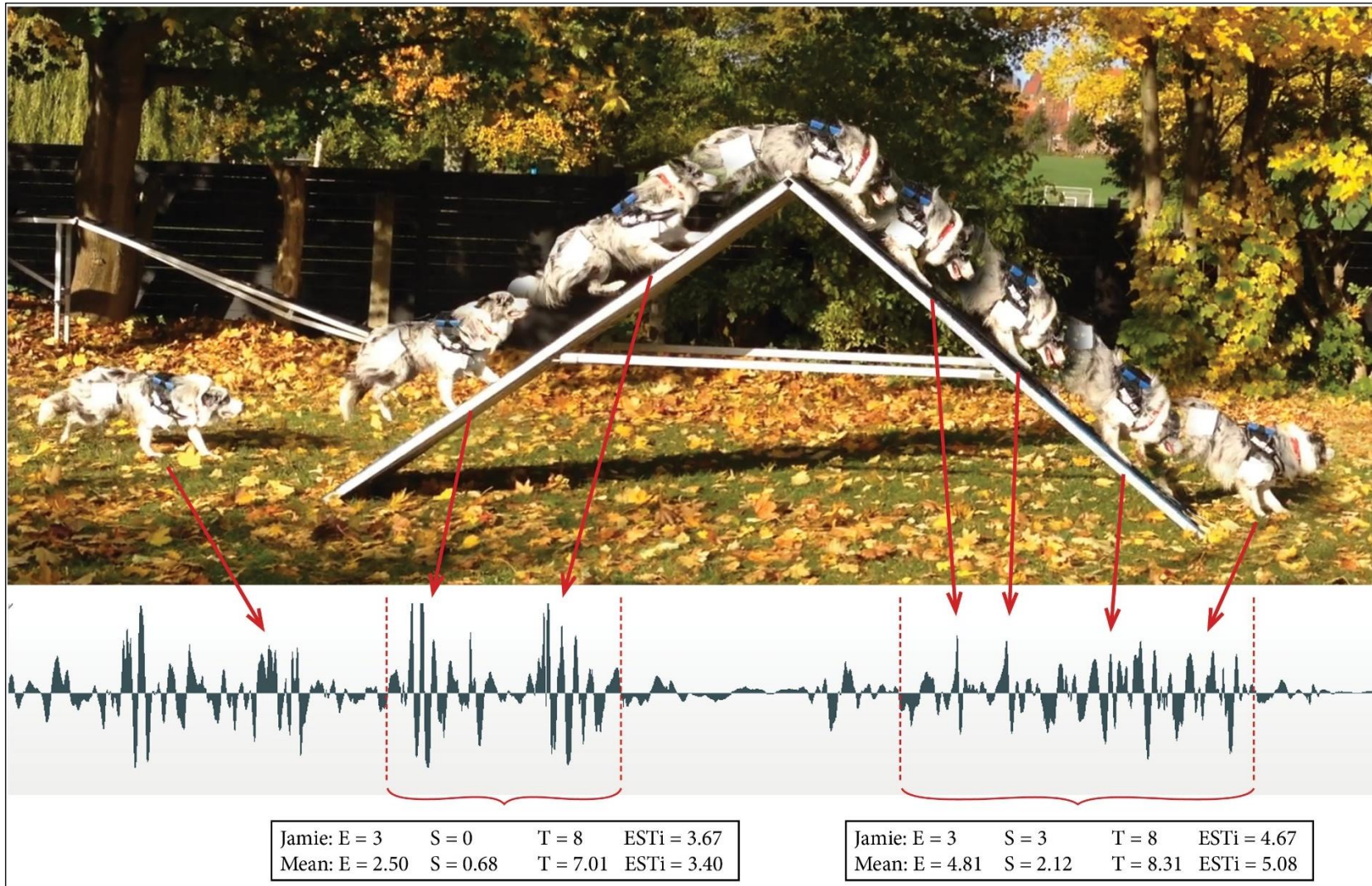
Table 15: Range and average for both amplitude and frequency in the two muscles triceps brachii and gluteus superficialis for dogs running a 170cm high A-frame practicing their usual contact method.

	Running up the A-frame		Running down the A-frame	
	Mean	Range	Mean	Range
<b>Triceps brachii</b>				
Amplitude (S)	5.5db	1.8-6.0db	4.7db	1.8-6.0db
Frequency (T)	35.9hz	0-108hz	20.3hz	0-84hz
<b>Gluteus superficialis</b>				
Amplitude (S)	4.7db	1.8-6.0db	2.41db	0.6-6.0db
Frequency (T)	20.9hz	0-48hz	30.7hz	0-120hz

#### 4.4.4 Conclusion

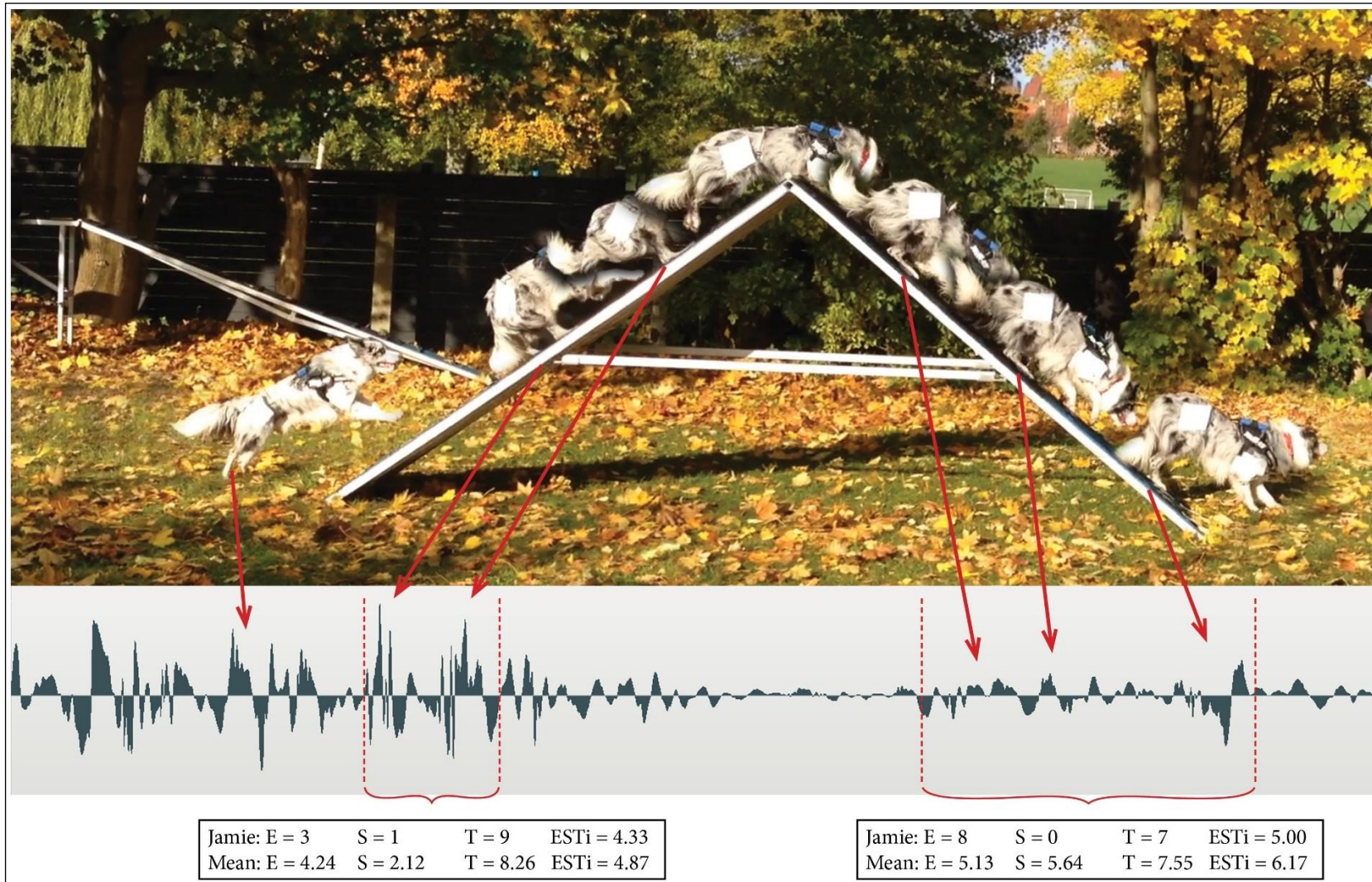
The shift from practising the 2o2o-contact method to the running contact method is likely to increase the risk of injury as well as repeated training on the A-frame. Changing the height of the A-frame to 120 cm and practising running contacts may also increase the risk of injury.





Picture 31: Illustration of a border collie (Jamie) running the A-frame practicing 2on-2off contact method compared with the recorded acoustic myographic signal of triceps brachii. Sections for analysis are indicated with the dashed red lines. The ESTi™-scores for Jamie in illustrated run and the average of 14 dogs running the A-frame three times are listed together with standard deviation (SD). The pictures are taken with an Iphone4 or 5 camera explaining the poor quality and the pictures should be interpreted carefully.





Picture 32: Illustration of a border collie (Jamie) running the A-frame practicing 2on-2off contact method compared with the recorded acoustic myographic signal of gluteus superficialis. Sections for analysis are indicated with the dashed red lines. The ESTi™-scores for Jamie in the illustrated run and the average of 14 dogs running the A-frame three times are listed together with standard deviation (SD). The pictures are taken with an Iphone4 or 5 camera explaining the poor quality and the pictures should be interpreted carefully.





	Up Jamie (mean)	Down Jamie (mean)
E	3 (2.50)	3 (4.81)
S	0 (0.68)	3 (2.12)
T	8 (7.01)	8 (8.31)
ESTi	3.67 (3.40)	4.67 (5.08)



	Up Jamie (mean)	Down Jamie (mean)
E	3 (2.65)	5 (3.98)
S	0 (0.62)	0 (2.79)
T	7 (7.18)	9 (7.74)
ESTi	3.33 (3.48)	4.67 (4.83)



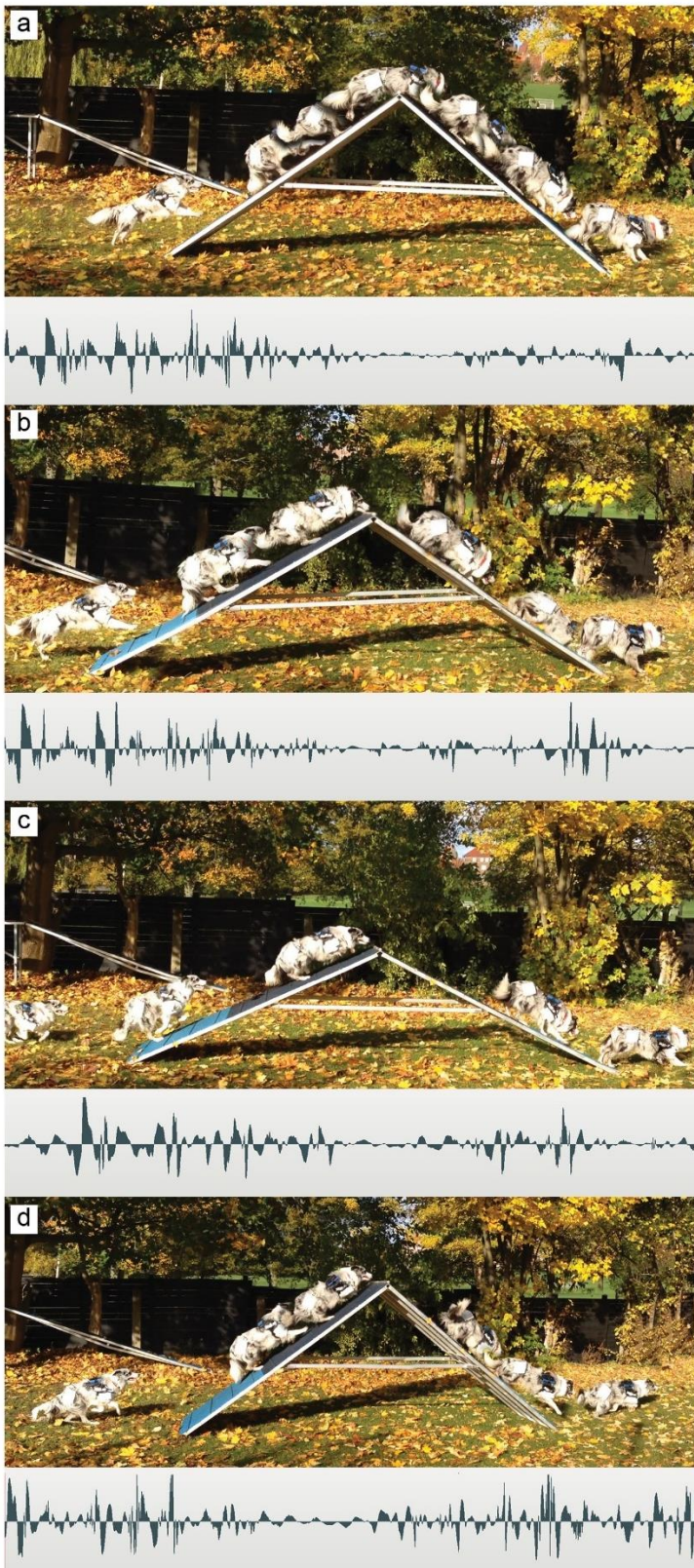
	Up Jamie (mean)	Down Jamie (mean)
E	7 (2.90)	4 (3.85)
S	2 (0.79)	0 (2.12)
T	10 (7.21)	8 (8.13)
ESTi	3.33 (3.61)	4 (4.70)



	Up Jamie (mean)	Down Jamie (mean)
E	1 (2.00)	1 (2.89)
S	0 (0.69)	2 (1.77)
T	8 (6.88)	9 (7.87)
ESTi	3.00 (3.19)	4 (4.18)

Picture 33: Illustration of a border collie (Jamie) running the high (h=170cm, picture a), medium (h=150cm, picture b) and low (h=120cm, picture c) A-frame using 2on-2off contacts method and running the high A-frame using running contact method (picture d). Each is compared with the recorded acoustic myographic signal of triceps brachii. The ESTi™-scores for the run up and down the A-frame respectively are listed for Jamie together with the average of 14 dogs running the A-frame three times. The pictures are taken with an Iphone4 or 5 camera explaining the poor quality and the pictures should be interpreted carefully.





	Up Jamie (mean)	Down Jamie (mean)
E	3 (4.24)	8 (5.31)
S	1 (2.12)	0 (5.64)
T	9 (8.26)	7 (7.55)
ESTi	4.33 (4.87)	5 (6.17)

	Up Jamie (mean)	Down Jamie (mean)
E	5 (5.12)	7 (4.60)
S	3 (2.05)	0 (5.88)
T	8 (8.19)	0 (7.04)
ESTi	5.33 (5.12)	2.33 (5.84)

	Up Jamie (mean)	Down Jamie (mean)
E	5 (4.57)	5 (4.80)
S	2 (2.45)	2 (6.30)
T	9 (7.70)	9 (7.43)
ESTi	5.33 (4.91)	5.33 (6.17)

	Up Jamie (mean)	Down Jamie (mean)
E	4 (4.17)	3 (4.21)
S	0 (1.67)	2 (5.14)
T	6 (7.76)	9 (7.52)
ESTi	3.33 (4.53)	4.67 (5.63)

Picture 34: Illustration of a border collie (Jamie) running the high (h=170cm, picture a), medium (h=150cm, picture b) and low (h=120cm, picture c) A-frame using 2on-2off contacts method and running the high A-frame using running contact method (picture d). Each is compared with the recorded acoustic myographic signal of gluteus superficialis. The ESTi™-scores for the run up and down the A-frame respectively are listed for Jamie together with the average of 14 dogs running the A-frame three times. The pictures are taken with an Iphone4 or 5 camera explaining the poor quality and the pictures should be interpreted carefully.

## 4.5 The cool-down exercise

The cool-down had no effect on neither triceps brachii nor gluteus superficialis. It was hypothesised that triceps brachii and gluteus superficialis showed a higher ESTi<sup>TM</sup>-score after the cool-down exercises partly due to the wash out of protons and reduction of acidosis (Robert *et al.* 2004). It is possible that the dogs did not perform an exercise intense enough to cause fatigue. Meanwhile only few signs of fatigue were seen during the previous trials suggesting that maximal capacity was not reached and a rehabilitation to normal level did not occur.

As mentioned in section 1.7.4 *The cool-down exercises*, there do not seem to be a difference in the ESTi<sup>TM</sup>-score when comparing horses walking before and after exercise. Differences was found when comparing the ESTi<sup>TM</sup>-scores before the warm-up exercises with the ESTi<sup>TM</sup>-scores after the cool-down exercises. A two-tailed paired sample t-test was used for triceps brachii and a Wilcoxon signed ranks test for paired samples with two tails for gluteus superficialis due to the data not being normal distributed. The S-score was significantly ( $p < 0.01$ ) higher after the cool-down exercises for both triceps brachii (pre-warm-up: 3.4, post-cool-down: 4.8) and gluteus superficialis (pre-warm-up: 5.6, post-cool-down: 6.9), while no other ESTi-parameter showed a significant difference. See result of test in appendix N. Thus, the dogs recruited fewer muscle fibres when trotting after approximately 1 hour of agility and cool-down exercises compared to when coming to the training area. Opposite the results for the 6 horses, the condition of the muscles in the dogs did not go back to normal condition, as the use of them where still improved. Does this mean they were ready for more exercises? A significantly ( $p < 0.05$ ) decreased E-score could be observed in triceps brachii when comparing the ESTi<sup>TM</sup>-scores after 15 minutes of warm-up exercises ( $E = 5.3$ ) with the ESTi<sup>TM</sup>-scores after the cool-down exercises ( $E = 3.1$ ). This suggest that triceps brachii was not in an optimal condition to work after the cool-down exercises, but gluteus superficialis was as no difference in the ESTi<sup>TM</sup>-score was found.

Although the cool-down exercise had no effect on the muscular tissues it may help the handlers to get control of their emotions after the run at competitions, and it is a good time to analyse and reflect upon causes of failure and success (Karvonen 1992). Hence cool-down exercises may also function as a mental cooldown.

### 4.5.1 Descriptive data

Dogs above 55cm had a higher firing rate in triceps brachii and generally used it less coordinated before the cool-down exercises compared with dogs below 55cm, but no difference was found between the two groups after the cool-down exercises. Dogs above 55cm were found to be more challenged when running up the high and medium A-frame resulting in an increased firing rate, and it is possible that the muscles is affected by this when trotting right after the trials with the A-

frame. The 15 minutes of cool-down reduces the difference between the tall and lower dogs, but without resulting in a significant difference when comparing the results before and after the cool-down exercises.

An effect of training level was seen and will be further discussed in section 4.6 *Training level*.

#### 4.5.2 Risk of injury

The performance of cool-down exercises did not influence the use of triceps brachii and gluteus superficialis and do not seem to reduce the risk of injuries. Meanwhile, a less coordinated use of triceps brachii after an hour of training is seen suggesting further training could increase the risk of injury. This would not have been detectable without the use of AMG as the dogs appeared fine and ready for more work.

If the taller dogs had continued the agility-trials before the cool-down exercises they could have been in increased risk of injury compared to the lower dogs as their muscle coordination was less efficient.

#### 4.5.3 Conclusion

The hypothesis of triceps brachii and gluteus superficialis showing a higher ESTi<sup>TM</sup>-score as a result of the performance of cool-down exercises is rejected.

### 4.6 Training level

Dogs training one hour a week or more had one or several increased ESTi<sup>TM</sup>-scores and hence improved use of triceps brachii compared to dogs training less than one hour a week during all exercises. Meanwhile no significant differences were found for gluteus superficialis. It is important to train on regular basis, preferably weekly, to improve the use of triceps brachii, while not doing so could increase the risk of injury in the forelimb which is already susceptible to injury (Levy *et al.* 2009; Cullen *et al.* 2013a, Cullen *et al.* 2013b).

Dogs training weekly recruited less muscle fibres in triceps after 10 and 15 minutes of warm-up exercises compared to dogs training less than one hour a week. This could indicate that warm-up exercises are more effective in well-trained dogs or that well-trained dogs do not need as long a period of warm-up exercises as less trained dogs to optimize their use of triceps according to recruitment of muscle fibres. They also recruited less muscle fibers in triceps brachii and were more effective and generally better coordinated both before and after cool-down exercises compared to dogs with less than one hour of training a week. It is possible that the training level had an influence on fatigue. When comparing the ESTi<sup>TM</sup>-score before and after the cool-down

exercises with the ESTi<sup>TM</sup>-score after 15 minutes of warm-up exercises for each group, conducting a Wilcoxon signed ranks test for paired samples with two tails, no significant differences were found for dogs training weekly (appendix N). Meanwhile the E and ESTi-score were significantly decreased (E and ESTi:  $p=0.02$ ) during the one-hour training for dogs training less than one hour a week. Weekly training improves the endurance of dogs performing agility, and not doing so may increase the risk of injury in the dogs as they easier get fatigued.

Cullen *et al.* (2013a) found dogs that trained less than one time a week to be in a higher risk (1.1 odds ratio) than dogs practising one time a week or more, although it was not significant in the final multivariable logistic regression model. Similarly, Zachary *et al.* (2014) found a decreased risk of injury when training more than 2 hours weekly being either agility training or walking. Dogs training less than one hour a week may be in a poorer fitness. Pope *et al.* (1999) found fitness to be a strong predictor of risk of injury in 1317 male army recruits. On the contrary Cullen *et al.* (2013a) found that dogs participating in conditioning exercises, such as aerobic, strengthening, proprioceptive or balance training, had a higher risk of injury (1.1 odds ratio) although the difference was not significant. It could be recommended to avoid strengthening-exercises as dogs use type II fibres when jumping (Sjaastad *et al.* 2010), which is found to be reduced in humans doing heavy-resistance-training (Staron *et al.* 1991), and this may be the same for dogs. Doing condition exercises in young beagles resulted in a higher percentage of type I fibres in triceps, again reducing the percentage of type II fibres, which is unwanted. Still, an increase of type II fibres was found in the lumbar multifidus muscles. Bruusgaard *et al.* (2010) recommend to practise strength training or give steroid treatment in humans to produce as many myonuclear in the muscle fibres as possible before aging as satellite cell activation is impaired in the elderly. While the percentage of type II muscle fibres may be lowered when training, it shall be noted that the number of myonuclear increases, also for type II fibres. Short but intense interval-training such as sprinting results in an increased percentage of type II muscle fibres (Dawson *et al.* 1998; Sjaastad *et al.* 2010). It can be recommended to do both endurance and strength training (Sjaastad *et al.* 2010), especially before the dog reaches an age where the ability of exercise-induced hypertrophy is reduced and the percentage of type II fibres decreases (Braga *et al.* 2016).

Dogs training less than one hour a week could be more prone to injury due to low fitness resulting in fatigue. Of course, the risk of agility related injuries would be none when not training at all, and with more time spent on training agility the risk would presumably increase as there is a risk of getting injury of 1.74 injuries per 10000 runs during competition and 1.72 injuries per 10000 hours during training (Zachary *et al.* 2014).



Meanwhile not only the training level, but also agility experience of both handler and dog has an influence on the risk of injury. Dogs having more than 4 years of experience have lower odds of injury and similar when the handlers have more than 5 years of experience (Cullen *et al.* 2013a). Furthermore, experienced dogs take-off and land further away from the hurdle at greater speed when compared to less experienced dogs (Birch *et al.* 2015), suggesting that experienced dogs have a better jumping technique. In the current study, experience had no effect, but this could be due to the division at 1 year of experience in contrast to the other studies. A more optimal comparison could be done having more dogs with more than 4 years of experience. Thus, it is not the first year of training that makes the difference.

Knowing this it could be recommended to take particular care in the first years of practice to avoid injuries, especially because dogs that have previously been injured are more susceptible to injuries (Jonhagen *et al.* 1998; Brockett 2004; Cullen *et al.* 2013a). Additionally, it is recommended to practice weekly to improve endurance and avoid muscle fatigue and injuries especially in the forelimb.

## 4.7 Daphne

No dog had identical results and the graphs could visibly differ much between the dogs, but one dog was remarkably different. Daphne, an old English sheepdog, had od AMG recordings and ESTi™-scores which is visualised in picture 36 where the AMG recordings from Daphne are compared with the recordings from a border collie (My). Picture 36 illustrates how the ESTi™-scores for triceps brachii is generally higher for Daphne compared to My. This is also true when comparing with the average of all the dogs for both muscles when trotting after 15 minutes of warm-up exercises and jumping, but no general pattern is seen when running the A-frame. During the trials, it was informed that Daphne has hip dysplasia<sup>2</sup> degree D, but with no symptoms according to the owner. It was decided to implement the dog in the trials as it does represent dogs practising agility, which is the population this sample is taken from and must represent. It is not uncommon that dogs with hip dysplasia in different degrees continually practise agility, and it is heard that some owners first knew of the condition after years of practise. Despite this, when having one dog with hip dysplasia out of 14 participants, dogs with hip dysplasia are probably over-represented in this study. The odd data could likely be due to the disease, but Old English Shepherds has a very long and dense coat and Daphne is no exception. A long, dense coat may

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<sup>2</sup> Hip dysplasia is a progressive structural change of the hip joint resulting in joint laxity, femoral head subluxation, deformity of the head, erosion of the articular cartilage and the development of degenerative joint diseases such as osteoarthritis (Alexander 1992; Oberbauer *et al.* 2017). The dogs are graded on a scale from A to E, where A is healthy dogs and E is dogs with severe hip dysplasia (Brass & Durán 2000; Durán 2017).

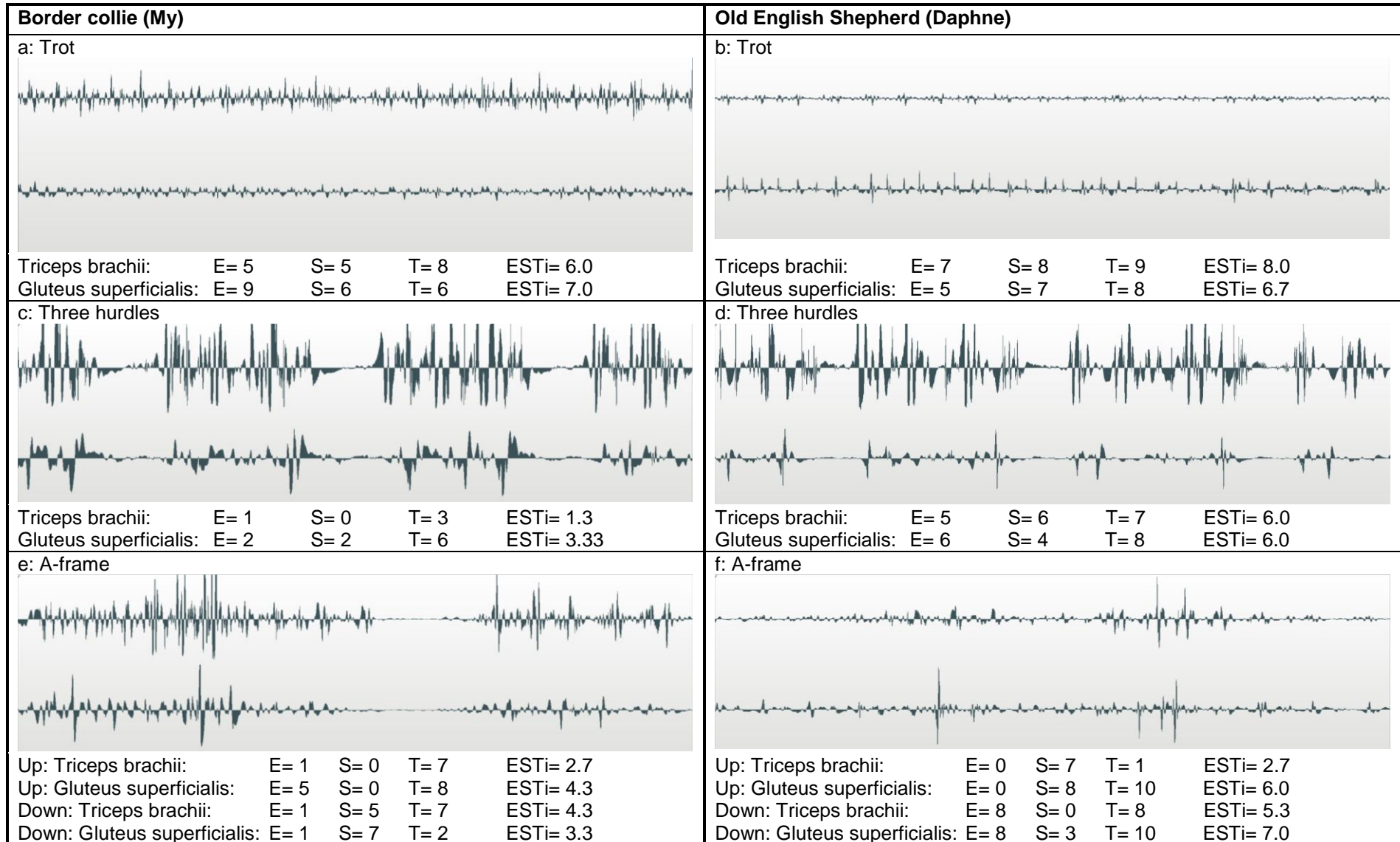


influence the measurements with the AMG as the sensors may not reach the skin (Fenger 2017) which could also explain the low AMG recordings seen in picture 36b and f. On the contrary, when looking at picture 36d this did not seem to be the case, in fact the Snøgg was firmly fixed in the fur as visualised in picture 35.



Picture 35: Illustration of how Snøgg adheres to the long dense coat of Daphne an Old English Shepherd, after she has been doing 10 minutes of warm-up exercises.

The well-being of Daphne, according to the possibility of pain related to performing agility, could be of concern when letting a dog with hip dysplasia perform agility. Meanwhile if Daphne felt pain during the trials the T-score would be low as a higher firing rate is seen when pain is associated with the use of the muscle (Graven-Nielsen *et al.* 2000; Vaegter & Graven-Nielsen 2016; Harrison 2017b). The T-scores were generally higher or near the average for all the dogs in both muscles. Only when running up the high A-frame for the second and the third time a low T-score (T=1) is seen in triceps brachii, but not when running down. Meanwhile her S-score is increased, this suggests that the higher firing rate compensate for a lower recruitment of muscle fibres giving the power needed to run up the A-frame. Additionally, her T-score is noticeable higher when jumping hurdles, no matter the height. Especially triceps brachii is used more efficient and coordinated with a lower firing rate and less muscle fibres recruited when comparing with the average of all the dogs. This suggests that Daphne does not associate the performance of agility with pain and it is possible she has learned to use her front-limbs more efficient to compensate for the hip dysplasia.



Picture 36: Illustration of differences between a healthy border collie and an Old English Shepherd with hip dysplasia degree D. Acoustic myography recordings and ESTi™-scores are shown for each dog trotting after 15 minutes of warm-up exercises (a,b), jumping three 55 cm high hurdles (c,d) and running up and down a 170 cm high A-frame (e,f). Data can be seen in appendix J, K and L.

## 4.8 Improvements

Besides the improvements mentioned in section 4.1 *Sources of error* other improvements could be applied.

Triceps brachii and gluteus superficialis was chosen due to both the function of the muscles, the increased risk of injury at the site, as well as practical reason according to the applied AMG-equipment. With the new OdB sensors it would not be a problem measuring the full amplitude of triceps brachii, but with the sensors used in this study, the use of another forelimb muscle could have been considered. In the pilot study trapezius was first considered, but due to concerns about the position of the sensors close to the harness trapezius was avoided. Additionally, the possibility of measuring an effect from the height of the hurdles and A-frame was of concern as several more distal muscles were expected to take most of the increased impact. Nonetheless as most of the agility related injuries occur in the shoulder region (Levy *et al.* 2009; Cullen *et al.* 2013a) the effect on this muscle could have been interesting to measure. The second most common site was the back of the dogs, but no further definition was given of the site of injuries. It could be interesting measuring different back-muscles of the dog, to find the cause of the increased risk and which muscles the handler, veterinarian, chiropractor, masseur etc. should be aware of and give special care.

It could be interesting measuring the effect on smaller dogs, and most dogs in the medium class, which are 35-42.9cm high at the withers (Federation Cynologique Internationale 2017a), could probably run with the CURO on their back. Still, the weight of the CURO would consist of a higher percentage of the dogs' body mass with risk of altering their moving pattern (Carrier *et al.* 2008).

It is evident how similar the muscle activation occurs when comparing low with high hurdles and with a high-resolution video camera synchronized with the CURO and CURO Equine-program a detailed analysis of the muscle activation would be possible. With the recording made in this thesis only qualified guesses can be made although it is possible to synchronize the AMG-recordings with the videos to some extent as done in the pictures 27, 28 for the hurdles and 31, 32, 33 and 34 for the A-frame. Much can be learned from the combination of AMG and video recordings and deeper investigations in real-time muscle activation and use could be done.

There are improvements to this study and most encourage to further studies implementing the improvements or giving alternative approaches.

## 5.0 Conclusion

This study is the first of its kind to use AMG to investigate how triceps brachii and gluteus superficialis in dogs is affected when performing three different heights of hurdles and the A-frame as well as 15 minutes of warm-up and cool-down exercises. It has been shown that:

- 1) The use of triceps brachii and gluteus superficialis does not improve with decreased hurdle height, on the contrary they use their muscles more efficient and coordinated when jumping hurdles with a height of 65 cm. Dogs weighing above 21 kg recruits fewer fibres than dogs weighing 21 kg or less when jumping hurdles above 55 cm.
- 2) A lower A-frame results in a less efficient and coordinated use of triceps brachii, but had no effect on gluteus superficialis. Opposite, dogs practising the 2o2o-contact method recruits less muscle fibres in gluteus superficialis. Additionally, triceps brachii showed an increased firing rate for dogs with a height above 55 cm at the withers running up an A-frame higher than 150 cm.
- 3) Dogs practising running contact method did not have an improved use of the muscles compared to dogs practising the 2o2o-contact method. On the contrary, practicing the 2o2o-contact method improved the efficiency and overall coordination in triceps brachii when running up the A-frame, and similar findings were seen when running down for both muscles. Dogs used to practice the 2o2o-contact method were less efficient and coordinated when shifting to practicing running contact method.
- 4) The use of triceps brachii improves after 5 minutes of warm-up exercises, but an effect of warm-up exercises was not found for gluteus superficialis.
- 5) Fifteen minutes of cool-down exercises had no effect on triceps brachii and gluteus superficialis.

Dogs training one hour a week or more improve their use of triceps brachii and do not fatigue as quickly as dogs training less than one hour a week. Dogs with hip dysplasia do not necessarily feel pain when performing agility and are able to adapt to the conditions.

Risk of agility-related injuries can be reduced, when knowing that dogs may be in increased risk of injury when jumping low hurdles, running low A-frames and shifting from practising the 2o2o-contact method to running contact method. Doing warm-up exercises and training at least one hour a week may as well reduce the risk of injuries.

Further studies are required to determine the ideal height of the A-frame and hurdles as well as the optimal duration of warm-up exercises. The results may inspire agility organizations, instructors and dog handlers to re-evaluate the current practise of agility.

## 6.0 Perspective

Some effect of altered height of the hurdles and the A-frame has been shown in this thesis and this can be implemented in training and competition to minimize the risk of injury in dogs performing agility. The results have been compared with previous studies investigating the risk of injury and doing so agility has been portrayed as a sport with a high risk of injury which preferably should be avoided by dog lovers. Meanwhile no studies have compared the risk of injury in dogs performing agility with regular family dogs not participating in any canine sports. When participants for the experiments was first recruited, 17 dogs were entered, but before executing the experiments three dogs were excluded due to injuries. One was due to a collision with another dog, the second due to a regular walk in the morning and the third because the dog jumps up and down with excitement in the daily life. Thus, although there are risks when participating in agility there is also risks in the daily life and no relative comparison has been made. It is possible that dogs participating in agility are less prone to injuries in the daily life as an improved coordination and general use of the muscles could be expected. A study investigating how dogs participating in canine sports use their muscles compared to untrained family dogs could be done with the use of AMG and a survey of whether or not the risk of injury is reduced when participating in canine sports could be interesting.

The thesis has shown some results in how to avoid injuries and knowing this it is possible for the stakeholders to take advantage of this. The agility organizations and the FCI may use this as an argument to alter or keep the regulations, as many participants ask for lower heights of the hurdles and the A-frame which may not be in their best interest. The trainer may also use the knowledge to educate the handlers in the increased risk of injury and how to avoid it. The handler can also implement the findings in their training by always performing at least five minutes of warm-up exercises before beginning the training. They also know that it may not be a good idea to lower the hurdles or A-frame with the purpose of allowing more runs before the dogs get fatigued, as this would merely make the dog use its muscles less coordinated and increase the risk of injury. Instead the handler should let the dog practice on the normal height and only practice on the A-frame three times during training. It is simply quality before quantity. When deciding which contact method to use, the results in this thesis have favoured the practice of the 2o2o-contact method. The handler must also be aware of the different challenges and advantages when jumping hurdles and running the A-frame according to the height of the dog.

The results may also be implemented in other canine sports such as flyball, rally-obedience, the DcH-program, utility dogs and rescue dogs. Flyball is a relay race between two teams of four dogs.

Each dog jump over four hurdles, retrieve a ball and run back over the four hurdles (Federation Cynologique Internationale 2016). The fastest team wins and the sport requires a high speed and accuracy from the dog. In this sport, a warm-up exercise could be implemented as perfect jumping technique and effectiveness is fundamental from the first hurdle and weekly training should be implemented. In flyball milliseconds divide the winning and the losing team, unlike agility where the margin is broader and has another focus. In this sport, a perfect jumping technique is also required, but the regulation cause some restrictions. The height of the hurdles is positioned according to the length of ulna in the smallest dog. Thus, if a lower dog enters the team the hurdle height may be lowered and without the necessary training of jumping techniques this will increase the risk of injury according to the results in this thesis. It could be recommended to have fixed heights and that dogs in same heights participate together avoiding the risk associated with changing of the hurdle height.

Hurdles and alternative A-frames are used in rally-obedience<sup>3</sup>, the DcH-program<sup>4</sup> and for utility dogs<sup>5</sup> (Danmarks civile Hundeførerforening 2017c; Danmarks civile Hundeførerforening 2017b; Federation Cynologique Internationale 2011c). Meanwhile common for all of them is that the sizes of the obstacles do not change and the dog shall only learn one running or jumping trajectory and can do so to perfection. Additionally, the dogs only have to jump the obstacles once or twice during a competition, thus no injury due to repetition happens. The hurdles are only 40cm high for large dogs performing rally-obedience, which in this study has shown to alter the dogs jumping techniques making them less careful and coordinated. Rally-obedience is not a sport in which the

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<sup>3</sup> In rally-obedience the dogs must perform different obedience and agility-related exercises explained by signs positioned on a course (Danmarks civile Hundeførerforening 2017c). One of the tasks is to jump one or two subsequent hurdles. The hurdles are 15, 25 or 40 cm high and positioned according to the height of the dog.

<sup>4</sup> In the DcH-program, which is an obedience program, an obedience-hurdle is used which shall not be mistaken for an agility hurdle as it is a smaller version of an A-frame being only 1meter high (Danmarks civile Hundeførerforening 2017b). Here the dogs must run over the obedience-hurdle fetch an object and then run back over the obedience-hurdle to the handler.

<sup>5</sup> The canine sport "utility dogs" also called IPO (Internationale Prüfungs-Ordnung) consists of exercises related to the work as police dogs. They must jump 1meter high hurdles and climb a 1.8 meter high A-frame with an angle under apex at 40° (Federation Cynologique Internationale 2011c). For comparison, it is 100° for agility.



dog shall perform physically, but it could be recommended to heighten the hurdles to demand a proper jumping technique from the dogs and thus avoid injuries. Meanwhile dogs do not necessarily gallop, but merely walk or trot to the hurdle thus affecting the whole jumping trajectory. The dogs may not use the elastic recoiling effect as efficient, thus jumping higher hurdles would be demanding and recruitment of muscle fibres or a higher firing rate could be necessary. It could be suggested to draw the course in such a way that the dogs are allowed sufficient space to assess the hurdles in their preferred pace. Opposite the above mentioned canine sports rescue dogs do not have a fixed height of the hurdles or potential ramps they shall climb when: jumping up and down stable and unstable planks, jumping into cars or boats, running planks and ladders, although some measurements are shown with approximate measures in the tests (Federation Cynologique Internationale 2011b). With the results from this thesis it is recommended that these dogs shall be trained weekly and trained in all heights and angles to achieve an effective jumping trajectory. The handler should as far as possible ensure a warm-up period and good circumstances to make the jumps or climb the ramps when in work. This could be done by offering the dog enough space to judge the obstacle and enter it at a preferred gait. Similar for all the canine sports is that they could benefit from implementing warm-up exercises as both physical and obedience canine sports could benefit from an improved neuromuscular coordination.

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## Appendix

Appendix A: Agility obstacles

Appendix B: Facebook posting

Appendix C: E-mail to participants

Appendix D: Description of location of the training area

Appendix E: Short description of the trial

Appendix F: The protocol

Appendix G: Contract between dog owner and Lene Høeg Callesen

Appendix H: The questionnaire

Appendix I: Body condition score

Appendix J: Data for the warm-up exercises

Repeated measures analysis of variance

Data and tests for the warm-up exercises

Appendix K: Data and tests for jumping the hurdles

Appendix L: Data and tests for running up the A-frame

Data and tests for running down the A-frame

Appendix M: Data and tests for the cool-down exercises

Appendix N: Extra tests