

Institutionen för husdjursgenetik

A genetic study of summer eczema in Icelandic horses

by

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Examensarbete 282 2006

Examensarbete ingår som en obligatorisk del i utbildningen och syftar till att under handledning ge de studerande träning i att självständigt och på ett vetenskapligt sätt lösa en uppgift. Föreliggande uppsats är således ett elevarbete och dess innehåll, resultat och slutsatser bör bedömas mot denna bakgrund. Examensarbete på D-nivå i ämnet husdjursgenetik, 20 p (30 ECTS).



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Agrovoc: Horses, genetics, heritability, culicoides Övrigt: Summer eczema, insect hypersensitivity

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1. Referat

Studier har visat att flera hästraser runt om i världen uppvisar en överkänslighet mot bitande insekter. Hästar har visat sig var mer känsliga mot vissa specifika insekter än andra. *Culicoides* är ett av de släkten som är mycket omskrivet då det gäller denna överkänslighet hos häst. Reaktionen mot insekterna visar sig i huvudsak som klåda, vilket kan leda till att hästarna kliar sönder sin hud och sår kan uppkomma. I Sverige och i övriga Europa har detta uppmärksammats hos flera raser och vad gäller Islandshästen är det mycket omdiskuterat. Det har visat sig att de hästar som exporterats från Island i vuxen ålder har en högre sjukdomsprevalens då de inte har haft någon chans att bygga upp ett immunförsvar mot insekten eftersom denna inte förekommer på Island. Även en viss andel av hästar födda utanför Island är drabbade av sjukdomen, som i Sverige benämns som sommareksem.

Målet med denna studie var att skatta genetiska parametrarna för graden av sommareksem hos svenskfödda islandshästar i Sverige. Denna studie bygger på enkätsvar som samlats in från islandshästägare i Sverige under sommaren 2005. Enkäten sändes ut till ägare till svenskfödda avkommor efter 33 utvalda hingstar. Resultatet bygger på information om 825 hästar.

Totalt 8% av hästarna var drabbade av sommareksem. Ston med eksem hade en högre andel avkommor som också var drabbade av eksem, jämfört med friska ston. Arvbarheten skattades till $0,20 \pm 0,1$ med en far modell och $0,19 \pm 0,1$ med en djurmodell. De fixa effekterna som användes i modellen var hästen ålder och hästens geografiska lokalisering i landet. Ålder hade ingen signifikant betydelse (p> 0,05) medan hästens lokalisering tycks vara av betydelse (p< 0,05) för om hästen utvecklar eksem eller inte.

2. Abstract

Several horse breeds around the world have shown sensitivity to biting insects. The horses seem to be particularly sensitive to certain species of the genus *Culicoides*. The main reaction to the insects' bites is itchiness and some horses itch until wounds appear. In Sweden and the rest of Europe many breeds have been observed to have the problem and as regards Icelandic horses this phenomena is widely discussed. It has been shown that horses exported from Iceland to continental Europe as adults have higher disease prevalence. These horses have less chance to develop a normal immune defense to *Culicoides*, since no *Culicoides* have been found in Iceland. Nevertheless, a certain proportion of Icelandic horses born outside Iceland are also affected by the disease, which in Sweden is known as summer eczema.

The aim of this study was to estimate genetic parameters for severity of summer eczema in Icelandic horses born in Sweden. The study is based on information from a questionnaire sent to owners of Icelandic horse in Sweden during the summer of 2005, with questions regarding the status of their horses' eczema. The questionnaire was sent to owners of Swedish born horses after 33 selected stallions. The result is based on the information about 825 horses.

A total of 8 % of the Swedish born horses were affected with summer eczema. Affected dams were more likely to have offspring with eczema, compared with healthy dams. The heritability was estimated at 0.20 ± 0.1 using a sire model and at 0.19 ± 0.1 using an animal model. The fixed effects included in the model were age and geographic location of the horse at present time. Age did not seem to be of significance (p>0.05) for the trait, whereas geographic location on the other hand showed to be significant (p< 0.05) for whether the horse develops eczema or not.

3. Introduction

The horse industry is a growing sector in the Scandinavian countries. Iceland has the largest number of horses per capita in Scandinavia: 280 000 inhabitants and 80 000 horses (Embassy of Iceland, 2005). The Icelandic horse of today origins from horses brought to the island by the Vikings and the Irish. They were in need of small and strong horses, and only the most durable survived the journey over the Atlantic sea. Due to the isolated location of Iceland, and a law from 900 A.D. that prohibits bringing horses into the country, the old Viking horses' gaits and character are maintained. The northern countries of Europe have also discovered the qualities of these animals and the population is rapidly increasing, both in number of horses born in these countries and through regular import of horses from Iceland (Eidfaxi, 2005).

When export of horses from Iceland increased, it was noticed that many horses developed difficulties with itching in their new countries, especially around mane, tale and hind areas (Troedsson and Broström, 1986). The disease, usually referred to as summer eczema, causes suffering for the horse and also economical loss for the owner (Riek, 1953). Icelandic horses born outside of Iceland can also develop the condition, but the frequency is lower compared with Icelandic born horses (Broström *et al.*, 1987). It has been shown that the major reason for this disease is an allergic reaction to protein in the saliva of the biting insects belonging to the genus *Culicoides*. This gnat was reported not to exist in Iceland by Hesselholt and Agger (1977), and we have found no reports in the literature of any findings during recent years.

In February 2005 there were 18 098 registered Icelandic horses in Sweden. Of these, 56% were born in Sweden and 44% were imported. Almost 90% of the imported horses were born in Iceland (SIF register, 2005). The total number of horses exported from Iceland in 2004 were 1 578 (Eidfaxi, 2005), and 450 of these were imported to Sweden (Eidfaxi, 2004). Summer eczema is an important issue for horse breeders in Iceland, who want to sell their horses abroad. The export has decreased with more than 1000 horses per year since 1996 and one of the reasons might be the risk for the exported horses to develop summer eczema in their new countries (Eidfaxi, 2004).

3.1 Aim of study

This report is part of a larger project that investigates the genetic background of summer eczema in Icelandic horses, and the candidate genes that might be involved.

The aim of this study was to estimate genetic parameters for severity of summer eczema in Swedish born Icelandic horses in Sweden.

4. Background

4.1 History

Summer eczema is the term used in Sweden (Broström *et al.*, 1987), Denmark (Hesselholt and Agger, 1977), Norway (Halldórsdóttir and Larsen, 1991a) and Germany (Unkel *et al.*, 1986) for the seasonal dermatitis that has been documented in horses in many parts of the world. Different names have been used to describe the condition such as "sweet itch" in England (Mellor and McCaig, 1974), "Queensland itch" in Australia (Riek 1953a), "kasen" in Japan (Nakamura *et al.*, 1956) and "summer sores" in France (Henry and Borey, 1937). Parallels have been drawn between countries worldwide, but it is not clear whether the different conditions described has a common or related origin. Sometimes similar clinical features, but with different origin, might be diagnosed as summer eczema (Quinn *et al.*, 1983; Halldórsdóttir and Larsen., 1991a; Frey, 2005).

The first recorded observation of summer eczema was in France in 1840 (Henry and Borey, 1937). In Australia the earliest records of the disease are from 1888 (Riek et al., 1953a). At that time the cause of the disease was not known. It is now clear that the seasonal eczema is caused by hypersensitivity to biting insects, but along the way other reasons have been suggested. Sunlight has been suggested to be the cause partly because the disease occurs during warmer weather, mostly on the dorsal part of the body, and horses kept inside seem to recover rather quickly. However, it is known from humans that hair is a very good protection against sunlight, and since the horses are mostly itching around the mane and tail area it was concluded that sunshine was unlikely the reason (McCaig, 1975). Moreover, this disease must not be confused with photo sensibility, which is a reaction between photodynamic substances in the skin and UV-light in unpigmented areas of the body (Broström, 2006). Grass has also been considered as a possible cause, but normally food allergy affects the whole of the skin area and not just certain parts as in the case with summer eczema. A small experiment has shown that affected ponies kept indoors while fed with newly cut grass did not show signs of the disease, whereas control ponies kept outdoors were still affected. Bacteria, fungi and lice have also been discussed, but no connection between fungi or bacteria and eczema horses has been reported in the literature. Lice commonly attack the mane area but are known to be killed by the rise in temperature during the spring, whereas summer eczema affects horses during summer time when the weather is warm. The evidence speaking for microfilariae as the reason are rather weak since the prevalence of affected horses with microfilariae is similar to the frequency of unaffected horses with microfilariae (McCaig, 1975).

Riek (1953b) was first to come to the conclusion that the disease may be caused by hypersensitivity to biting insects. Circumstantial evidence now declares that summer eczema is a hypersensitivity reaction to the saliva of any of several species of biting insects of the genus *Culicoides* from the family *Ceratopygnidea* (Riek, 1954; Yamashita *et al.*, 1957; Quinn *et al.*, 1983; Troedsson and Broström, 1986; Anderson *et al.*, 1988; Fadok and Greiner, 1990).

4.2 Insect activity

More than 90 species of the genus *Culicoides* are known and none of them has been observed in Iceland (Hesselholt and Agger, 1977; Illies, 1978). The gnats are small and their larvae have been identified in mud, sand, and debris at the edge of ponds, springs and creeks (Logas and Barbet, 1999). Only the female *Culicoides*, which have larger salivary glands than males, feed on blood (Wilson *et al.*, 2001). The occurrence of these insects is considered to be closely related to the weather (Nakamura *et al.*, 1956). Adult gnats are most active when there is little or no breeze, and the temperature exceeds 10° C (Logas and Barbet, 1999). Nakamura et al. (1956) found that the degree of itchiness in the horse runs parallel with the rise and fall of atmospheric temperature, and the most severe itchiness was observed after a rain period followed by clear weather.

During the most active hours of *Culicoides*, several hundreds can be caught from a single horse in one hour (Wilson *et al.*, 2001). Different species of *Culicoides* have different life cycles with respect to active hours and seasons. Riek (1954) found for example, when investigating horses in Australia, that *C. robertsi* are never observed (with rare exceptions) biting horses during the middle of the day. They are present in greatest number before 7 a.m. and between 6 p.m. and 10 p.m. These times coincide with the time when the horses seem most troubled by itchiness. Similar results have been found in Japan (Nakamura *et al.*, 1956). Two other species, *C. marksi* and *C. papalis*, have been observed to be active throughout the day.

Different species of *Culicoides* prefer different landing (feeding) sites on the horse and this could explain why horses in different parts of the world seem to be troubled by lesions on different parts of the their bodies (Mellor and McCaig, 1974).

4.3 Clinical signs and affected individuals

Summer eczema is a chronic, seasonally recurring and non-infectious dermatitis (Quinn et al., 1983). The clinical signs of summer eczema can look very similar to the signs of other allergens (Frey, 2005). Lesions are usually confined to the dorsal region of the horse and especially around the long hair parts (Riek, 1953a; Nakamura et al., 1954; McCaig, 1975; Ouinn et al., 1983). However, Fadok and Greiner (1990) showed that the majority of horses with seasonal eczema have signs both dorsally and ventrally. A study on horses in Canada showed that the most frequently affected area of the body was the ventral midline (Anderson et al., 1988). In severe cases, the lesions extend down the side of the horse and also to the face and legs. Itching is the most obvious clinical sign, and can lead to hairless spots and wounds (Riek 1953a). Chronic cases can result in the skin not recovering over winter. It stays thickened, dry, rough with hairless spots, and these cases can show a yearly gradual progression in severity of the clinical signs (Broström et al., 1987; Kleider and Lees, 1984). The horse's general state of health is normally not affected, although grazing might be affected due to constant itching. Body temperature remains normal and appetite is unimpaired (Riek 1953a). If the horse is severely itching, wounds can appear with risk of secondary infections (Halldórsdóttir 1990). Broström et al. (1987) found that horses born in Iceland have more serious lesions compared with horses born in Sweden.

Baker and Quinn (1978) observed that the disease is predominant in ponies but occurs also in larger horses, whereas Anderson *et al.* (1988) did not find any significant difference in frequency of summer eczema in ponies vs. horses. Reiher and Björnsdóttir (2004) stated that Icelandic horses do not seem to be more susceptible to summer eczema than other breeds, if they are born in the same environment. Anderson *et al.* (1988) found that 26% of horses and ponies in British Colombia showed sensitivity to biting insects. A study from Holland shows that 8 % of Shetland ponies are affected by insect hypersensitivity (Kapell, 2005). Ruyter (2005) found a corresponding frequency of 18% in Friesian horses in Holland. In a study of Icelandic horses exported to Denmark, Sweden and Germany, 35% were diagnosed with summer eczema (see Table 1). If more than two years had passed since the date of importation the frequency of affected horses was 50 % (Björnsdóttir *et al.*, 2004). The frequency of summer eczema among horses imported from Iceland to Norway, Sweden and Germany are significantly higher compared with the frequency among horses born in these countries (see Table 1).

(N=number of horses).			
Country of location	Ν	Horses born in Iceland	Horses born in respective
		(%)	country (%)
Sweden ¹	441	26.2	6.7
Norway ²	391	26.9	8.2
Germany ³	651	-	6.3
Germany, Sweden,	330	34.5	-
Denmark ⁴			

Table 1.Frequency of Icelandic horses affected with summer eczema in Sweden, Norway and Germany (N=number of horses).

¹ (Broström *et al.*, 1987)

² (Halldorsdóttir *et al.*,1991)

³ (Reiher and Björnsdóttir, 2004)

⁴ (Björnsdottir *et al.*, 2004)

A number of studies have shown that sex and colour bear no significance for susceptibility of the disease (Riek, 1953a; Hesselholt and Agger, 1977; Broström *et al.*, 1987; Anderson *et al.*, 1988; Halldórsdóttir and Larsen, 1991a). Braverman *et al.* (1983), however, found that stallions were more sensitive than mares and dark horses were more susceptible than pale horses. Nakamura *et al.* (1954) stated that the disease is unaffected by nutrition or breed. The prevalence of the disease varies with different geographic areas (Nakamura *et al.*, 1954; Broström *et al.*, 1987). For susceptible horses the conditions recur every year if preventive measures are not taken (Hesselholt and Agger, 1977; Anderson *et al.*, 1988).

Animals of all ages are susceptible to summer eczema, even though it is rarer for young animals to show clinical signs (Riek 1953a). The age at which horses first become affected can vary greatly, but the average age of onset seems to be between 2 and 6 years (Nakamura *et al.*, 1954; Anderson *et al.*, 1988; Reiher and Björnsdóttir, 2004). Number of grazing seasons is of greater importance for time of onset than age when looking at horses imported from Iceland. In imported horses the disease usually appears during the second to fourth grazing season (Broström *et al.*, 1987; Halldórsdóttir and Larsen, 1991a; Björnsdóttir *et al.*, 2004). A certain period of

sensitization seemed to be needed before susceptible horses develop eczema (Broström *et al.*, 1987).

In horses imported from Iceland, time of year for importation seemes to influence whether the animal develop eczema or not. Horses imported during a period of low insect activity seemed to run a higher risk of developing summer eczema compared to horses imported during the active insect period (Halldórsdóttir and Larsen, 1991a).

4.4 Preventive measures

Since Bancroft (1891) found that stabled horses with eczema might be spared from clinical signs of the disease, this has also been recognized by others. Riek et al. (1953b) showed that animals kept indoors between 4 p.m. and 7 a.m. from early spring until the following winter remained free from signs of the disease. When horses showing lesions while being kept outdoors were confined in stable during night, the disease disappeared within three weeks. However, clinical signs of the disease reappeared within three days after the animals were let outside again at night (Riek 1953b). These results were confirmed by Troedsson and Broström (1986). Many consider stabling horses during periods of *Culicoides* activity to be the best method of controlling the disease (Riek 1953b; Quinn *et al.*, 1983; Troedsson and Broström, 1986). It has also been shown that horses showing clinical signs of summer eczema at a certain geographic location can be fully recovered if moved to another location with less insect activity (Riek 1953a).

In a study of ponies in Sweden using the "anti-insect rug" (Figure 1), 80% of the owners experienced an obvious improvement in their pony's condition by using the rug (Lönell, 1996). Using an "anti-insect rug" on horses that are allergic to the gnat, or keeping the horse away from the insects in other ways for example by stabling, should be enough to keep the horse free from clinical signs of the disease. If these measures are not enough, the horse could possibly be sensitive to allergens other than *Culicoides* (Frey, 2005).



Figure 1. The anti-insect-rug is used to protect the horse from biting insects.

Treatment with antihistamines can give temporary recovery, but relapses seem to occur within a few days after the injections or withdrawal of oral treatment. Insect repellents sprayed sufficiently in parallel with antihistamine injections used

continuously for a long time can lead to complete recovery in some cases (Nakamura, *et al.*, 1957).

4.5 Immunology

Higher vertebrates have five classes of the antibody molecule immunoglobulin (Ig) and each has different roles in the immune response (Alberts *et al.*, 2002). The allergic reaction from the bites of *Culicoides* is an immediate immune reaction characterized by IgE antibodies release of histamine, leukotrienes and other inflammatory mediators (Sjaastad et al., 2003; Torsteindóttir 2004). Wilson et al. (2001) stated that IgE antibodies against Culicoides were only detected in the serum of horses with insect hypersensitivity but neither in the serum of normal horses exposed to *Culicoides* bites, nor in serum of recovered horses with a history of hypersensitivity. Similar magnitudes of IgG antibodies were detected from both allergic and healthy horses exposed to *Culicoides*. None of the antibodies were detected in Icelandic horses not exposed to *Culicoides* bites, i.e. horses living in Iceland. Larsen et al. (1988) suggested a conceivable theory to these findings: horses which are sensitized to the insect antigen during the first two to three summer seasons will produce both IgE and IgG. Horses producing mainly IgE may develop summer eczema and the ones producing mainly IgG will not show any signs of the disease. If a mare is producing enough IgG she will pass it on to her foal when nursing and the foal will run a lower risk of getting any clinical signs of the disease.

In humans and laboratory animals there is evidence that IgE responses to certain antigens can be specifically suppressed by passive transfer of maternal immunity via colostral milk from immunized mothers (Jarret and Hall, 1979, 1981). Icelandic mares in Sweden have been exposed to the allergens, whereas mares in Iceland have not. The lower prevalence of allergic dermatitis among horses born in Sweden may reflect passive transfer of maternal immunity to the foals by the IgG and IgA rich colostrum of their already sensitized mothers (Norcoss, 1982), that suppress subsequent development of hypersensitivity response to *Culicoides* as the foals mature under mosquito exposure. Thus, passive immunity may have a crucial role in regulating or modulating the switch of various isotypes of immunoglobulin and thus preventing development of allergic dermatitis later in life (Broström *et al.*, 1987).

Baker and Quinn (1978) and Quinn *et al.* (1983) challenged affected and normal horses with intradermal injections of extract from biting insects. When injecting *Culicoides* extract, made from the entire insect, into affected horses all reacted immediately and some also with a delayed reaction (after 24 hours). Extract injections from other insects (*Stomoxys calcitrans* and *Tabanidae*), or a saline control, did not give as strong reaction as *Culicoides*. When clinically normal horses were challenged with the extracts, they all showed minimal dermal reactions. By collecting serum from horses with active skin lesions present and by injecting it intradermally into unaffected horses, Quinn *et al.* (1983) showed that skin of clinically normal horses can be sensitized with serum from affected horses, i.e. sera from hypersensitivity horses contain IgE antibodies against *Culicoides* (Prausnitz and Kustner test). The skin thickness increased almost immediately after injection, and generally there was a close correlation between skin fold thickness and area of oedema in the recipient horse.

Halldórsdóttir and Larsen (1989) challenged 23 affected and unaffected Icelandic horses with extracts from four species of biting midges: *Culicoides pilicaris, C. chiopterus, C. obsoletus and C. impunctatus*. Ten of the 14 (71%) affected horses responded to at least three of the four antigens. Two of the affected horses did not show any response to either of the antigens, but these horses were very mildly affected by the disease. One of the nine unaffected horses (11%) responded to two of the extracts.

Fadok and Greiner (1990) tested the hypersensitivity of 44 horses in Florida with seasonal itchiness and 21 horses without symptoms with intradermal injections of allergic extracts from insects, grass and mould. The different insect dilutions were from *Culicoides*, mosquitoes, horse flies and black flies. The skin biopsies demonstrated changes compatible with arthropod hypersensitivity and the most intense reaction from the insect extracts were towards *Culicoides*. It was not possible to determine if reactions to a special insect were correlated with a specific clinical symptom. It was noticed that horses in general showed reaction to *Culicoides* alone or to *Culicoides* in combination with other insects.

MHC (major histocompatibility complex) is a complex of highly polymorphic genes that code for cell-surface glycoproteins that bind foreign proteins and peptide fragments and present them to T-cells to induce an immune response. In horses it is referred to as ELA and is located on the horse chromosome 6 (Alberts *et al.*, 2002). Marti *et al.* (1992) suggested that summer eczema is a multifactor disease including hereditary and environmental factors in its pathogenesis, since genes of the MHC appears to play a role in some families as well as genes outside the MHC. Halldórsdóttir *et al.* (1991b) tested horses exported from Iceland to Germany, Norway, Sweden, Denmark and Switzerland for their distribution of leucocytes antigens. Only one antigen gave a statistical significant difference in distribution, the Swiss specifity ELA-antigen, Be 8.

4.6 Heritability and variance components

Riek (1953a) observed that susceptibility to summer eczema may be related to the pedigree of the horse, and that there seem to be a higher correlation with the phenotype of the dam than the sire. McCaig (1975) also concluded that some horses seem to have a genetic predisposition for the disease and that affected horses are commonly found to have a parent or a grandparent with the disease. Björnsdóttir *et al.* (2004) found that the risk of getting affected was significantly higher for a horse if the parents (one or both) were affected with the disease. Unkel *et al.* (1986) concluded that the trait was influenced by common maternal environment. Marti *et al.* (1992) however did not find any association between dam and offspring, but agrees that susceptibility for dermal hypersensitivity due to biting insects most likely is hereditary.

In two studies of horses in the Netherlands heritabilities of insect hypersensitivity were estimated at 0.06 ± 0.02 and 0.06 ± 0.03 in Shetland ponies and Friesian horses respectively (Kapall, 2005; Ruyter, 2005). In these studies the horses were scored for summer eczema at foal inspections and eight inspectors did the scoring. The scale

used to identify the severity of the animals' condition was: 1) showed no insect hypersensitivity, 2) showed a few signs of insect hypersensitivity and 3) the animal was clearly affected. With more than 3000 horses recorded in each breed respectively, these are the largest genetic studies of the trait hypersensitivity to biting insects in horses.

In an earlier study Unkel *et al.* (1986) investigated the genetic basis of summer eczema in 984 Icelandic horses in Germany, and concluded that there is no monofactorial recessive or dominant inheritance. They estimated heritabilities for insect hypersensitivity between 0 and 0.24 ± 0.12 using different models and methods.

5. Material and methods

5.1 Data

A questionnaire was used to gather information from offspring of stallions having more than 50 offspring born in Sweden between 1991 and 2001. These criteria resulted in 33 stallions with a total of 3261 offspring born within the selected time period. The questionnaire was sent by letter to owners of the selected offspring, and owners of the stallions themselves, in the beginning of June 2005. Addresses were collected from the horse register at the Swedish Association of Icelandic horses, where most of the Icelandic horses in Sweden are registered. The owners were asked to fill out the questionnaire for all their horses, but it was emphasized in the letter that our major interest was to receive information about horses sired by the selected stallions. In this study only information from offspring of the selected stallions was used, and the information about other horses was saved for later studies. The questionnaire was also available on the internet and horse owners were primarily recommended to fill out this electronic version.

The questionnaire (see appendix A) recorded information about: the horse's identity, birth year, sex, country of birth, present location of the horse, severity of summer eczema, place and time of onset of summer eczema, time of year when the horse show clinical signs, affected areas of the body, other symptoms of allergy and information about possible clinical signs in the dam. Severity of summer eczema was recorded in four classes: 1=healthy (unaffected), 2=mild eczema (no clinical signs if preventive measures are taken), 3=moderate eczema (show itching even when preventive measures are taken). Preventive measures were, for example, stabling of the horse or use of an anti-insect rug, but not medical treatment. Three traits with different grouping of the four scores of eczema were created to establish which one was most useful for the genetic analysis.

Grouping of the scores of severity of eczema:

4 categories;	-	1=unaffected 2=mildly affected 3=moderately affected 4=severely affected
3 categories;	- - -	1=unaffected, 2=mildly affected 3=moderately and severely affected
2 categories;	- -	1=unaffected 2=mildly, moderately, and severely affected

Posters with information about the project were presented at the Swedish Championship and at the World Championship for Icelandic horses during the summer of 2005, where questions about the project could be answered. Articles about the project were published in the horse magazines "Islandshästen" and "Ridsport" to reach owners that may not have been reached by mail and as a reminder to all the selected horse owners. Since the questionnaire was available on an open access webpage, even people outside the selected group of horse owners could record information about their horses. However, only information from horses that fulfilled the selected criteria were included in the genetic analyses.

When we started to compile information for this study in September 2005, records from about 1660 horses were available. After data editing, records from 825 horses which were offspring of the selected sires, born during the selected time period and had complete information from the questionnaire remained. The total number of records received from the electronic questionnaire on the web-site was 583 (71%) and the rest, 242 (29%) were sent by regular mail. The total answering frequency was 27%. About 7% of the questionnaires were returned due to incorrect address information. The answering frequency among offspring from each stallion varied between 7% and 32%, with an average of 19% (Figure 2). The 825 horses with complete records had 652 dams, of which 32 had own observations for summer eczema in the data set. Of the dams, 123 had more than one offspring in the data set: 93 dams with two offspring, 21 dams with three offspring, 8 with four offspring and 3 dams with five offspring.

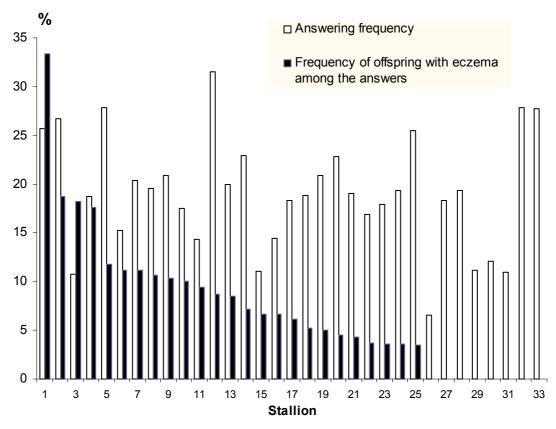


Figure 2. Answering frequency and frequency of allergic eczema in the offspring for the 33 stallions represented in this study (ordered by frequency of allergic eczema among the offspring in the study).

5.2 Statistical analyses

The data set was analysed using the SAS package (SAS Institute, Cary, NC, USA, version 8.2). PROC MEANS and PROC FREQ were used to get means and frequencies. To establish the influence of fixed effects PROC GLM was used. The horses were divided into six groups, depending on where in Sweden they were living at present. See appendix B for grouping, and Table 2. A single trait mixed linear animal model (1) was used to estimate variance components. To check for the influence of non-additive co-variation between dam and offspring, we also repeated the analyses using a sire model (2).

$$Y_{ijkl} = \mu + b_i + l_j + a_k + e_{ijkl}$$

$$\tag{1}$$

$$Y_{ijkl} = \mu + b_i + l_j + s_k + e_{ijkl}$$
⁽²⁾

where:

 $\begin{array}{ll} Y_{ijkl} &= \text{score of severity of summer eczema } (1, 2, 3 \text{ or } 4) \\ \mu &= \text{least square mean} \\ b_i &= \text{fixed effect of present age of the horse } (4 - 14, 11 \text{ classes}) \\ l_j &= \text{fixed effect of present geographic location of the horse } (6 \text{ classes}) \\ a_k &= \text{random animal effect } \sim \text{ND}(0, A\sigma_a^2), \text{ or } s_k = \text{random sire effect } \sim \text{ND}(0, A\sigma_s^2) \\ e_{ijkl} &= \text{random residual effect } \sim \text{IND}(0, \sigma_e^2) \end{array}$

The variance components were estimated using an average information (AI) restricted maximum likelihood (REML) algorithm (Jensen et al., 1997) in the DMU package (Madsen and Jensen, 2000).

6. Results

Of the total number of horses, 8% were affected by summer eczema (Figure 3). More than 50% of the affected horses had mild signs of eczema and fewer than 10% showed severe signs. The age of onset was 4.8 years (s.d. 3.0).

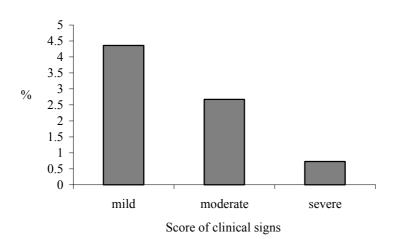


Figure 3. Frequency of the different scores of severity of summer eczema, as a percentage of the total number of horses

Present location of the horse was significant for severity of summer eczema. Table 2 shows differences between the regions (see appendix B for grouping of geographic regions).

Table 2. Differences	s between regions in severity of summer eczema, * = p<0.05, **=p<0.1, n.s.=non
significant, p>0.10.	The diagonal shows the means for the trait in each respective region.

	South	South-	South-	Mid-west	Mid-east	North
		west	east			
South	1.186	n.s.	*	*	n.s.	n.s.
South-west		1.209	*	*	**	**
South-east			1.071	n.s.	n.s.	n.s.
Mid-west		symm.		1.082	n.s.	n.s.
Mid-east		-			1.102	n.s.
North						1.085

Counties included in different regions: South - Skåne and Blekinge; South west - Västra Götaland and Halland; South east - Östergötland, Jönköping, Kalmar and Gotland; Mid west - Dalarna, Värmland and Örebro; Mid east - Gävleborg, Uppsala, Västmanland, Stockholm and Södermanland; North - Norrbotten, Västerbotten, Jämtland and Västernorrland.

The age of the horse was not significant for exhibition of the trait.

For 50% of the horses affected by allergic eczema, and 40% of the healthy horses in the data set, information about the condition of the dam regarding summer eczema was also available. Among offspring with dams reported to be affected with summer eczema, the frequency of eczema was higher compared with offspring to dams reported as healthy (22% compared with 7%) (Figure 4).

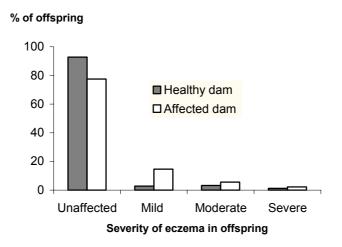


Figure 4. Clinical signs of allergic eczema in offspring of dams unaffected and affected by summer eczema, respectively.

The estimated variance components and heritabilities from the animal model are shown in Table 3 and from the sire model in Table 4.

Table 3. Variance components due to genetic effect of animal (σ_a^2) , random residual (σ_e^2) and phenotype (σ_P^2) , and heritabilities (h^2) , estimated with a linear animal model

Trait, eczema				
scored in:	h^2	σ^2_a	σ^2_e	σ^2_p
4 classes	$0.190_{0.089}$	0.039 _{0.019}	0.166 _{0.017}	0.205
3 classes	$0.126_{0.076}$	$0.021_{0.013}$	$0.148_{0.013}$	0.169
2 classes	$0.116_{0.074}$	$0.007_{0.005}$	$0.065_{0.006}$	0.072

Table 4. Variance components due to genetic effect of sire (σ_s^2), random residual (σ_e^2) and phenotype (σ_P^2), and heritabilities (h^2), estimated with a linear sire model

Trait, eczema				
scored in:	h^2	$\sigma^2{}_s$	σ_{e}^{2}	σ^2_p
4 classes	0.1960.095	0.0100.005	0.1940.010	0.204
3 classes	$0.119_{0.075}$	$0.005_{0.003}$	$0.164_{0.008}$	0.168
2 classes	$0.056_{0.058}$	$0.001_{0.001}$	$0.071_{0.004}$	0.072

The estimated heritability for the binary trait transformed to the underlying scale, according to Dempster and Lerner (1949) was 0.40 for the animal model and 0.19 for the sire model.

7. Discussion

7.1 Data

Studies have previously been done on Icelandic horses and their hypersensitivity to biting insects, but this is the first time genetic parameters have been estimated for the trait severity of summer eczema in Swedish-born Icelandic horses.

The answering frequency to the questionnaire was 27 %, which can be considered low. One problem when distributing the questionnaire was that the address information for some horse owners turned out to be incorrect. When buying a horse, it is up to the new owner to change the address in the register, but in many cases this has not been done and therefore the questionnaire and information about the project may not have reached all the selected horse owners. However, owners of selected horses who did not receive the questionnaire by mail may have read about the project in magazines and could still have answered the questionnaire on the web-site. The fact that there were no stamped returning envelop in the letter can also be a reason for the low answering frequency. The answering frequency for our questionnaire is, however, similar to the frequency of horses attending breeding shows, which are the ones the BLUP (Best Linear Unbiased Prediction) evaluation system for Icelandic horses is based upon today.

It is important to remember that the information used in this study was collected from horse owners. Owners of horses that have developed eczema are possibly more eager to answer the questionnaire than owners of unaffected horses, which could influence the results. The frequency (8%) of affected horses was, however, close to what has been shown in previous comparable studies of summer eczema (Table 1). The status of the horses was based on the owners own observation and that can of course vary from person to person depending on former experience and knowledge.

More than 70% of the owners used the internet based questionnaire. In the electronic version of the questionnaire, the compulsory information had to be filled out before one could move on. Consequently, the internet questionnaire yielded more complete records. The internet questionnaire, however, may have been more difficult to reach for people without computer skills.

We found the average age of onset to be, 4.8 years, which also agrees with what has been found in earlier studies (*eg* Broström *et al.*, 1987; Halldórsdóttir and Larsen, 1991a). However, previous studies have showen a widely spread distribution of the age when horses get affected. Differences between countries could be due to the fact that different species of *Culicoides* are present in different countries, and their antigen might affect the horses differently. There may also be differences between different breeds.

Our results indicate that affected dams are more likely to have offspring with eczema compared with unaffected dams (Figure 4). Similar results have been found in previous studies (Unkel *et al.*, 1986; Björnsdóttir *et al.*, 2004). This can be explained by hereditary factors, but it must also be considered that the dam and offspring spend some or sometimes all the time in the same environment, and under the same kind of treatment. In our data, 32 horses with eczema had recorded information regarding

clinical signs of eczema in the dam. About 50% of horses with eczema had information about the condition of the dam, compared with 40% of the unaffected horses. This difference could possibly reflect that owners of affected horses are more eager to search for information about their horse's background. Unfortunately, the question regarding the condition of the dam was only present in the internet based questionnaire, which of course contributes to the high frequency of missing information.

7.2 Heritabilities and environmental effects

The highest heritabilities (0.19-0.20) were estimated when severity of allergic eczema was scored in 4 classes. This is expected since we can utilize more variation in the trait by using a finer scale when scoring the trait. Therefore it is important to record summer eczema in a way that can use as much variation in the trait as possible. The heritabilities estimated in our study are higher compared with the results from two recent Dutch studies by Kapall (2005) and Ruyter (2005). These two studies used comparably large data sets. However, they only scored mares and inspectors judged the horses' eczema status, not the owners themselves. Previous studies on for example behaviour traits have shown that information from owners of animals have yielded higher heritabilities compared with information retrieved using tests at a single occasion (Grandinson et al., 2003). Even though the inspectors have potentially seen more horses and may be more objective, they only got to see the horse at one occasion. A well managed eczema horse can appear free from clinical signs, and that makes it difficult for an inspector to accurately score the disease. The owner is more aware of the "true" severity of the eczema since they know how much work is needed to keep the horse in a certain condition. If owners score their horses, influence of environmental factors such as weather, season and any preventive care, can be reduced. The fact that our study and the studies by Kapall (2005) and Ruyter (2005) were done on different breeds can of course also influence the heritability. In a German study on Icelandic horses, Unkel et al. (1986) estimated heritabilities in the range 0 - 0.24 depending on the model used. The higher estimates were estimated from models and methods more similar to the ones in our study.

Our estimated heritability of 0.20 shows that genetic progress in this trait might be possible, using information that can be provided by horse owners. The heritabilities estimated in this study are higher than what we expected, given the large environmental influence on the development of the disease and the categorical nature of the trait. Even if we can utilise more variation in the trait by recording in several classes, the most accurate way of analysing summer eczema would be to use a threshold model that accounts for the non-normal distribution of the trait. However, analyses using threshold models have often proven to be more sensitive to data structure and small datasets, and considering the limited size of our data we decided on a linear analysis. Transformation of the heritability from the binary analysis to the underlying scale shows a considerable amount of genetic variation that we do not see even when using the scale with more categories. Accurate estimates of genetic parameters and breeding values will require information from a larger number of animals, and with a larger data set a threshold model might be a better choice for the genetic analysis.

Several authors have previously suggested a maternal influence on the development of summer eczema (Riek, 1953a; Unkel *et al.*, 1986). Because of our limited data set we were unable to include a maternal component in our models. To check for influence of non-additive variation between dam and offspring, we instead analysed the trait using both a sire model and an animal model. A maternal influence would then be expected to over estimate the heritability estimated from the animal model, compared with the sire model where the dams are not included in the pedigree file. However, the estimated heritabilities were very similar between the two models when several classes of eczema were analysed, and we can therefore assume that any maternal influence is neglegible for this trait. The animal model can then be considered a better choice since it uses more pedigree information.

Icelandic horse stallions affected by summer eczema are taken out of breeding since the animal welfare legislation in Sweden forbids breeding with animals having a condition that is not considered healthy. This law applies only to stallions, since they get more offspring than mares. Stallion owners are required to sign a contract guaranteeing that their stallions are not affected by summer eczema. It is not desirable to have unhealthy animals in the breeding stock, and stallions with eczema are obviously sensitive since they have developed the disease. However, it is important to bear in mind the large influence from environmental factors on this disease and consequently the difficulties with ranking potential breeding animals based on phenotype. We need a tool to help us rank breeding animals in a more accurate way.

Excluding affected stallions does obviously not provide us with a chance to rank unaffected stallions with respect to the disease. We have no information about possible insect sensitivity among apparently unaffected stallions, which are often kept in very controlled environments. The care of the stallions is often different from mares and geldings. In most cases, stallions are kept inside at night during the entire year, whereas mares and geldings are often kept outdoors day and night during spring, summer and autumn. By keeping possibly sensitive stallions away from the allergens they might escape clinical signs of the disease. A seemingly healthy horse could thus inherit the eczema to its offspring to a larger extent than an affected horse. Putting all stallions together in the same enclosed pasture in an environment with high *Culicoides* activity for a season could provide us with information on which stallions who would develop eczema or not. However, this is obviously not ethically, economically or practically possible. A more practical way, that would also enable us to rank breeding mares, would be to estimate breeding values for the trait based on a BLUP animal model using records provided by horse owners.

A factor that is believed to be of large importance for whether the horse develops eczema or not is if the horse receives the important antibodies from its dam (Larsen *et al.*, 1988). The large difference in prevalence of summer eczema among Swedish born horses and imported horses is unlikely explained by different genetic predisposition for this trait between the two groups but rather a sensitization of susceptible adult horses to a new environment. Many of the breeding horses used in Sweden are imported from Iceland and thus never had a chance to develop an effective immunity as foals. It would thus not be fair to compare imported horses with horses born in Sweden. A genetic evaluation for summer eczema should therefore be based on horses born in Sweden. Imported horses would then get their breeding values based on information from Swedish born relatives.

With this data we were not able to study the effect of the owner of the horse. The owner is most likely of great importance for severity of summer eczema in an individual horse. If the owner is taking good care of an affected horse, it can in many cases appear completely free from clinical signs of the disease. The owner's choice, how to care for the horse, can of course also be essential for whether the horse develop eczema or not. Horses kept inside or carrying an "anti-insect rug" during the insects' active hours have a larger chance to escape eczema due to sensitivity to biting insects.

7.3 Recording and breeding strategies

Breeding value estimation with progeny testing for summer eczema, recorded as information from horse owners, will make it easier for breeders to choose the best animals to breed healthier horses. By using the BLUP methodology we can compare different animals and see who has the best genetic merit for the trait.

Summer eczema is a difficult trait to handle in a breeding programme. One major difficulty will be how to maintain updated reliable information about the horse population, since the condition in individual animals can change quickly over time. If a horse, for example, is scored at the age of four and at that moment does not have any clinical signs of eczema the horse will be recorded as healthy. If the same horse later develops the disease, the record needs to be updated. Geographic location of the horse would have to be corrected for in the breeding value estimation, and this information would also have to be updated if the horse is moved. Since the eczema has been reported in some studies to get worse with increasing age, age may also need to be corrected for in the model.

It could potentially be a problem to get all horse owners to accurately report the condition of their horses. For example, stallion owners that have an economic interest in their horse and in its offspring might not gladly reveal information about the stallion's condition, especially not since stallions with summer eczema immediately have to be taken out of breeding. One alternative would be to let veterinarians report if they encounter a horse affected by the disease. Even if clinical signs of the disease are not visible at all times of the year, this could be a step in the right direction. Recordings can also be done at breeding shows and competitions. At these events the owners always signs an insurance to guarantee that the horse is healthy. If a question whether the horse has eczema or not could be included, many horses eczema status could be recorded in that way. For competition horses, this is also a way to get updated information every time the horse competes. Some horses also attend breeding shows more than once. An internet based questionnaire, like the one used in this study, should be available for horse owners so that they may update information if their horse's eczema condition changes. In the end, it is all about getting healthier horses and most people would probably do their best to help make progress in the breed. A moral responsibility especially lies with the stallion owners and breeders.

Another important issue is how and when to publish the sensitive information that breeding values for summer eczema would be. Using the information collected for this study is a good start, however more data is needed before we can estimate breeding values with high enough accuracy. There will never be information available for the entire Icelandic horse population in Sweden, but the today existing breeding values used for confirmation and riding abilities are based on the scores from breeding shows where only around 30 % of the horses are represented. Future work of handling these problem lies with the breeding organization. Hopefully this information can be used to create the basis for a constructive discussion on selection methods for a healthier Icelandic horse population.

8. Conclusions

About 8 % of the Icelandic horses born in Sweden are affected with summer eczema, confirming that this is not a negligible problem in the Swedish born Icelandic horse population.

The heritability estimated in this study for the trait severity of summer eczema shows that it is possible to breed for healthier horses. The trait is under a rather large environmental influence, but by scoring severity of the eczema in several classes, instead of just healthy or affected, more genetic variation can be identified.

The fact that mother and offspring share a common environment does not seem to influence the results, and we found no evidence for non-additive maternal influence on the trait severity of summer eczema. We recommend an animal model to be used for analysis of this trait because it includes more complete pedigree information.

Since there is a large environmental influence on the trait, it is not enough to just look at the horse's phenotype when ranking breeding animals. By adding summer eczema as a trait to the breeding goal, we can use the BLUP evaluation system to help us rank our breeding animals in the same way as is done for conformation traits and riding ability. Progeny testing would be the best alternative to accurately handle horses imported from Iceland.

More horses have to be recorded to enable a more accurate and continuing estimation of genetic parameters for severity of summer eczema in the Swedish Icelandic horse population.

9. Future prospects

This report is a first preliminary result of this study. The questionnaire on internet closed in the beginning of the year 2006, since a new eczema season will start during the spring. New data will be added during the spring 2006 and genetic parameters will be re-estimated. The next step in the project is to choose families for DNA sampling, to find markers that can help us identify genes involved in hypersensitivity to biting insects.

Another interesting project would be to investigate the eczema frequency of the offspring born in Sweden from dams imported during pregnancy. The mare has not had any opportunity to give her offspring antibodies against biting insects, but the foal

is still born into this environment. How susceptible are these horses, compared to horses born in Sweden by Swedish born dams, or imported dams that have spent at least one grazing season in Sweden? How important are the antibodies that the foal receives from its mother for susceptibility to summer eczema? We have found no reports of such studies in the literature.

10. Acknowledgement

First I would like to thank all the owners of Icelandic horses that have answered the questionnaire and by that made this study possible.

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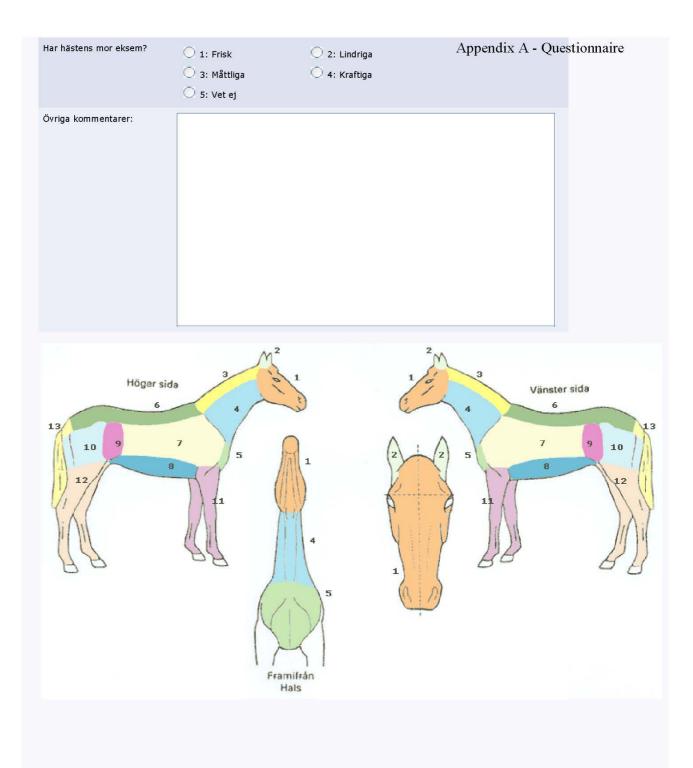
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	Uppgifter om häst	Appendix A - Questionna
Hästens fullständiga namn, inklusive gårdsnamn: *	[]	
FEIF-nr/reg.nr/passnr:		
	Oregistrerad	
Födelseår:	Vet ej	
Kön:	Hingst Osto Ovallack	
Hästens fader inkl. FEIF/reg.nr./stamboksnr.:		
Hästens moder inkl. FEIF/reg.nr./stamboksnr.:		
Födelseland:	Osverige Island Norge Danmark Tyskland Annat land	
Annat land, ange här:		
När importerades hästen?	År: Mån: Vet ej	
Huvudsaklig ort där hästen befinner sig nu: *		
Postnummer för den ort där hästen befinner sig: *		
	Uppgifter om eksem	
Hur grava är hästens problem? *	🔿 1: Frisk 🔷 2: Lindriga	
	🔾 3: Måttliga 🦳 4: Kraftiga	
Hur utbredda är hästens eksem? *		
3 1	1.Ansikte 2.Öron	
6 4	3.Man 4.Hals	
10 9 7 5	5.Bröstvirvlar 6.Korset/ryggen	
13 8 12 11	9.Flankvirvlar	
N N	11. Framben	
Klicka på bilden för förstoring	13.Svans	
Plats och tidpunkt för eksemut	brott: *	
Plats/ort:		
Postnummer:	Vet ej plats/ort/postnr	
Tidpunkt:	År: 📃 🗌 Vet ej år	
	Årstid: ○ Vår ○ Sommar ○ Höst ○ Vinter ○ Vet ej årstid	
När kliar sig hästen? *	🗌 Vår 🔲 Sommar 🔲 Höst 🗌 Vinter	
Andra allergisymptom?	Luftvägsbesvär Nässelutslag Annat Inte aktuellt	

ire





- Skåne, Blekinge
- Västra Götaland, Halland
- Östergötland, Jönköping, Kronoberg, Kalmar, Gotland
- 4. Gävleborg, Uppsala, Västmanland, Stockholm, Södermanland
- 5. Dalarna, Värmland, Örebro
- 6. Jämtland, Västernorrland, Västerbottan, Norbotten