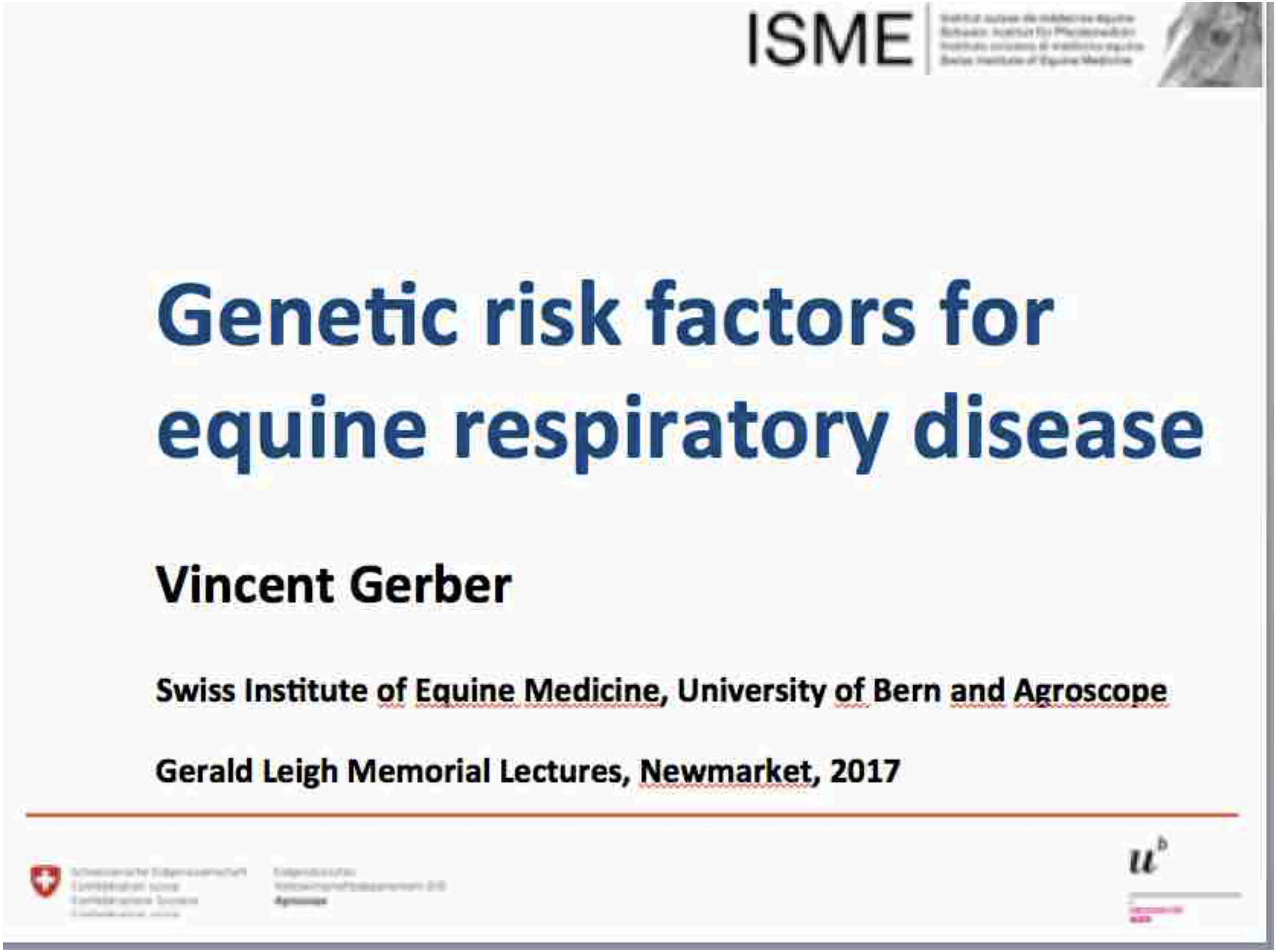


ISME

Genetic risk factors for equine respiratory disease

Vincent Gerber

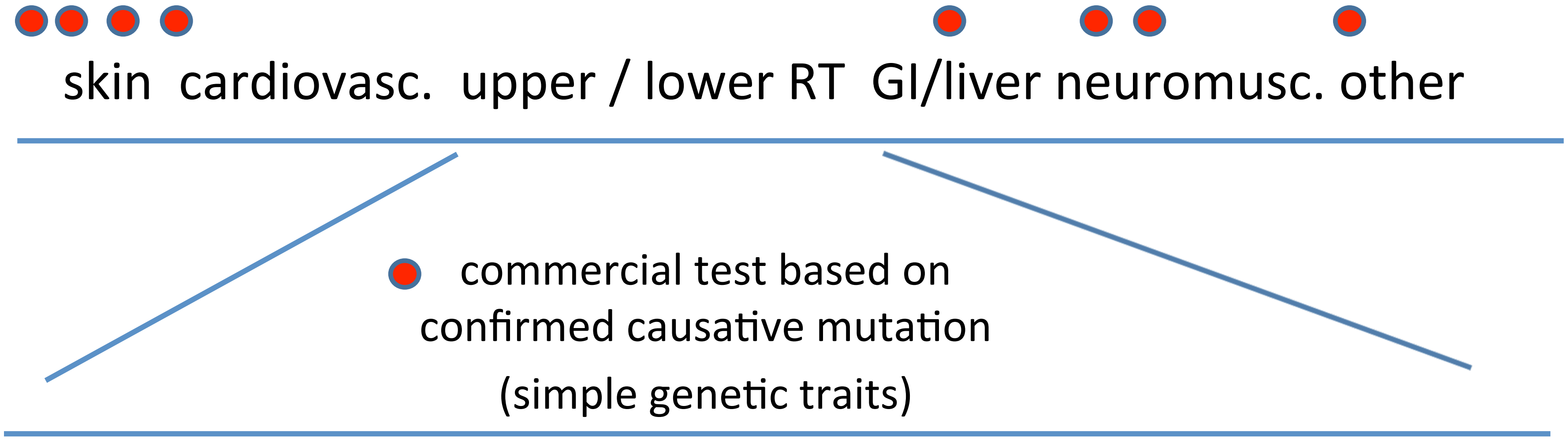
Swiss Institute of Equine Medicine, University of Bern and Agroscope

Gerald Leigh Memorial Lectures, Newmarket, 2017

“tour d'horizon”

- alar fold… wry nose, 4BAD …
- DDSP, pharyngeal instability / -collapse
- Guttural pouch tympany
- RLN
- Pharyngitis, Rhodococcal pneumonia, streptococcal lower airway disease
- EAV, EHV1/4, EHV5
- EIPH
- Equine asthma - IAD vs. RAO

Medical genetics of the horse by organ system & disorder



●● ●● ● ●● ●

skin cardiovasc. upper / lower RT GI/liver neuromusc. other

● commercial test based on confirmed causative mutation

(simple genetic traits)

alar fold… wry nose 4BAD,

DDSP, pharyngeal instability / -collapse

Guttural pouch tympany

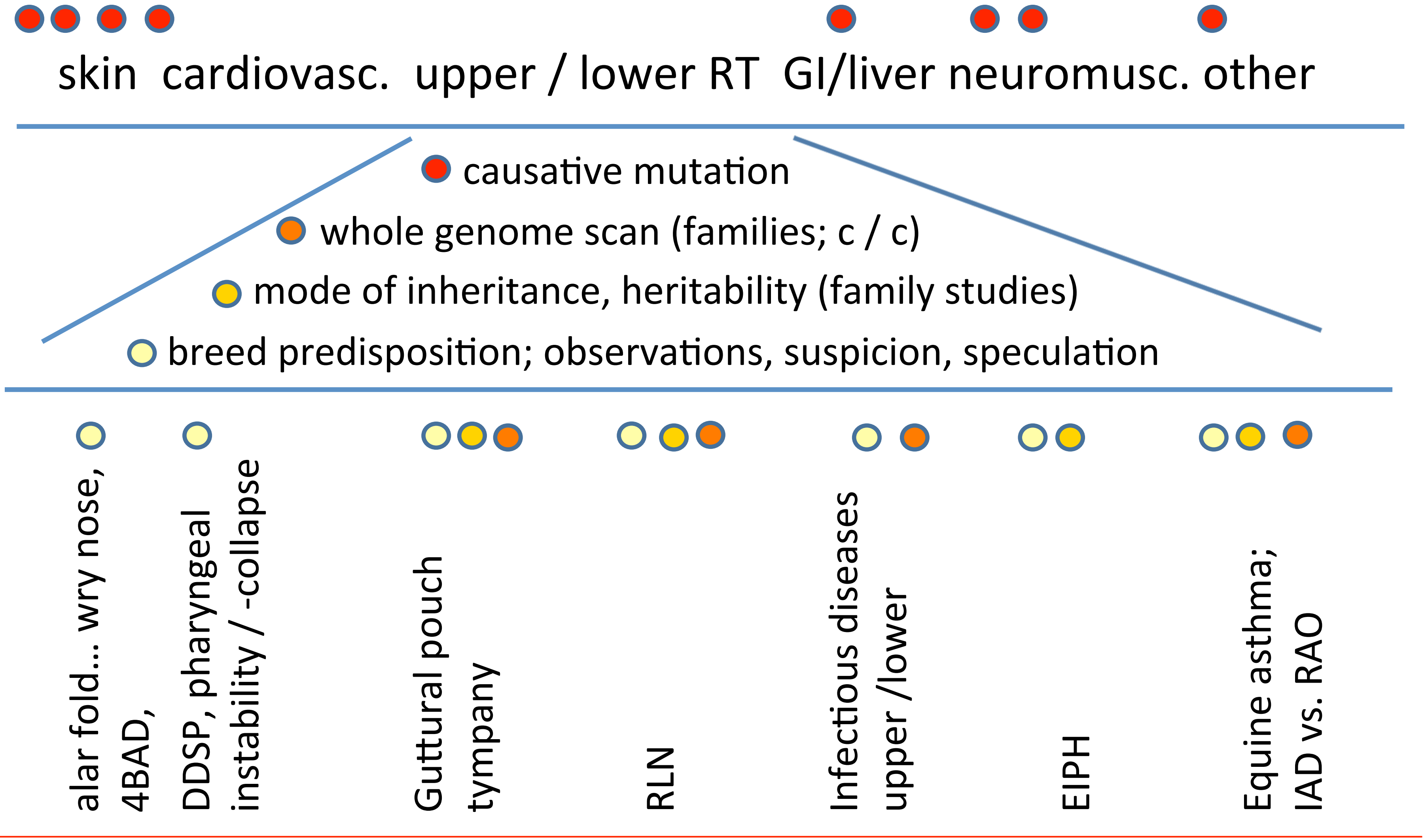
RLN

Infectious diseases upper /lower

EIPH

Equine asthma; IAD vs. RAO

Medical genetics of the horse by organ system & disorder



🔴🔴 🔴 🔴 🔴 🔴 🔴 🔴

skin cardiovasc. upper / lower RT GI/liver neuromusc. other

🔴 causative mutation

🟠 whole genome scan (families; c / c)

🟡 mode of inheritance, heritability (family studies)

○ breed predisposition; observations, suspicion, speculation

- ○ alar fold… wry nose, 4BAD,
- ○ DDSP, pharyngeal instability / -collapse
- ○ 🟡 🟠 Guttural pouch tympany
- ○ 🟡 🟠 RLN
- ○ 🟠 Infectious diseases upper /lower
- ○ 🟡 EIPH
- ○ 🟡 🟠 Equine asthma; IAD vs. RAO

tour d'horizon

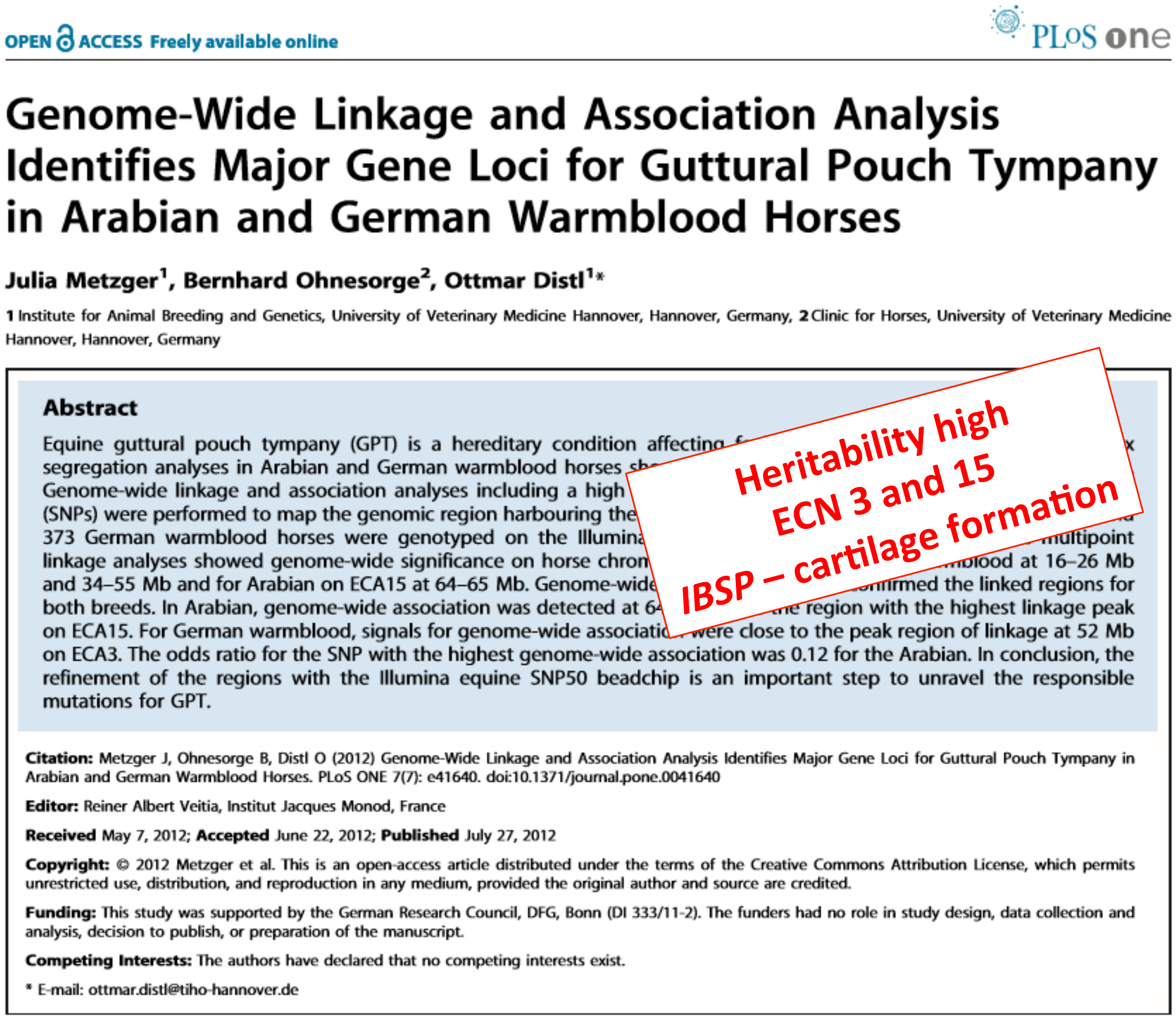
- alar fold… wry nose, 4BAD,
- DDSP, pharyngeal instability / -collapse
- Guttural pouch tympany
- **RLN**

- Pharyngitis, Rhodococcal pneumonia, Streptococcal lower airway disease
- EAV, EHV1/4, EHV5

- EIPH
- **Equine asthma** - IAD vs. **RAO**

Guttural pouch tympany

YES



OPEN ACCESS Freely available online

PLoS one

Genome-Wide Linkage and Association Analysis Identifies Major Gene Loci for Guttural Pouch Tympany in Arabian and German Warmblood Horses

Julia Metzger[1], **Bernhard Ohnesorge**[2], **Ottmar Distl**[1]*

1 Institute for Animal Breeding and Genetics, University of Veterinary Medicine Hannover, Hannover, Germany, 2 Clinic for Horses, University of Veterinary Medicine Hannover, Hannover, Germany

Abstract

Equine guttural pouch tympany (GPT) is a hereditary condition affecting x
segregation analyses in Arabian and German warmblood horses
Genome-wide linkage and association analyses including a high
(SNPs) were performed to map the genomic region harbouring the
373 German warmblood horses were genotyped on the Illumina multipoint
linkage analyses showed genome-wide significance on horse chrom at 16–26 Mb
and 34–55 Mb and for Arabian on ECA15 at 64–65 Mb. Genome-wide the linked regions for
both breeds. In Arabian, genome-wide association was detected at region with the highest linkage peak
on ECA15. For German warmblood, signals for genome-wide association were close to the peak region of linkage at 52 Mb
on ECA3. The odds ratio for the SNP with the highest genome-wide association was 0.12 for the Arabian. In conclusion, the
refinement of the regions with the Illumina equine SNP50 beadchip is an important step to unravel the responsible
mutations for GPT.

Citation: Metzger J, Ohnesorge B, Distl O (2012) Genome-Wide Linkage and Association Analysis Identifies Major Gene Loci for Guttural Pouch Tympany in Arabian and German Warmblood Horses. PLoS ONE 7(7): e41640. doi:10.1371/journal.pone.0041640

Editor: Reiner Albert Veitia, Institut Jacques Monod, France

Received May 7, 2012; **Accepted** June 22, 2012; **Published** July 27, 2012

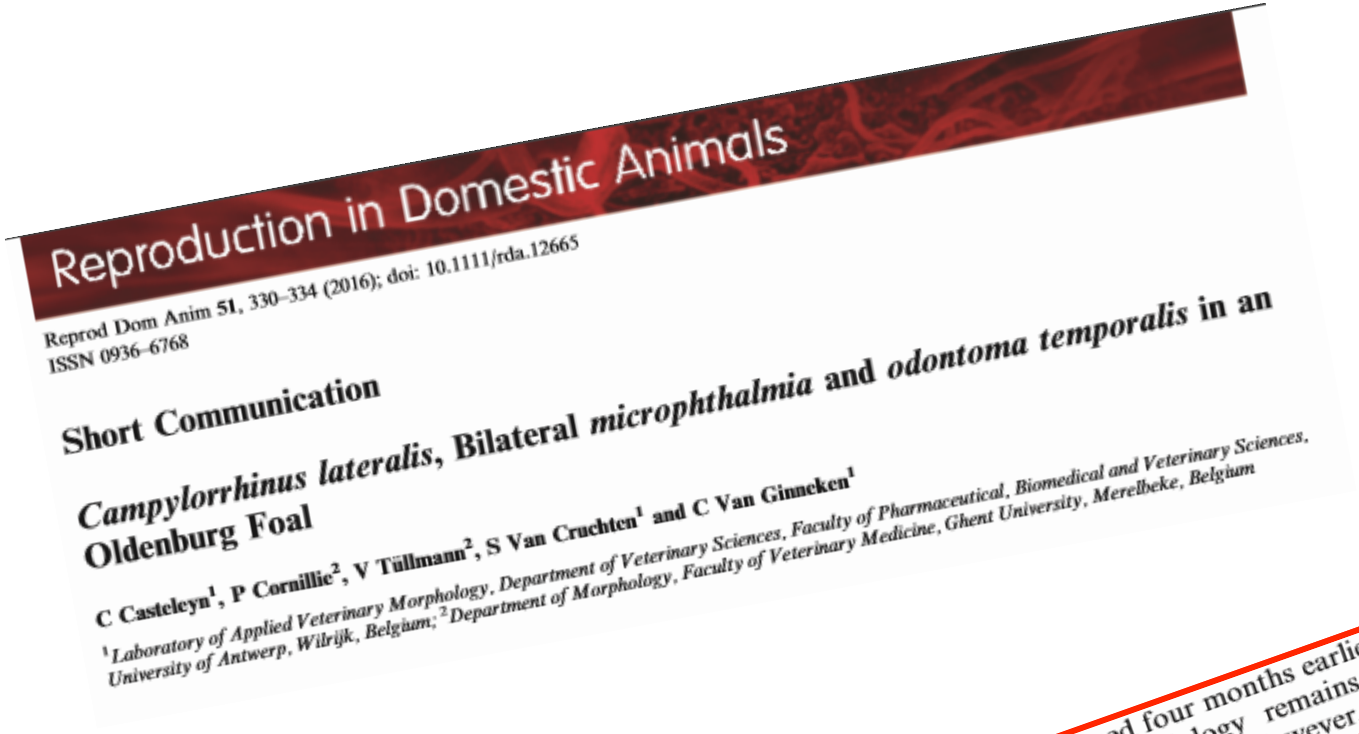
Funding: This study was supported by the German Research Council, DFG, Bonn (DI 333/11-2). The funders had no role in study design, data collection and analysis, decision to publish, or preparation of the manuscript.

Competing Interests: The authors have declared that no competing interests exist.

* E-mail: ottmar.distl@tiho-hannover.de

Heritability high
ECN 3 and 15
IBSP – cartilage formation

Birth defect – low heritability


Reproduction in Domestic Animals

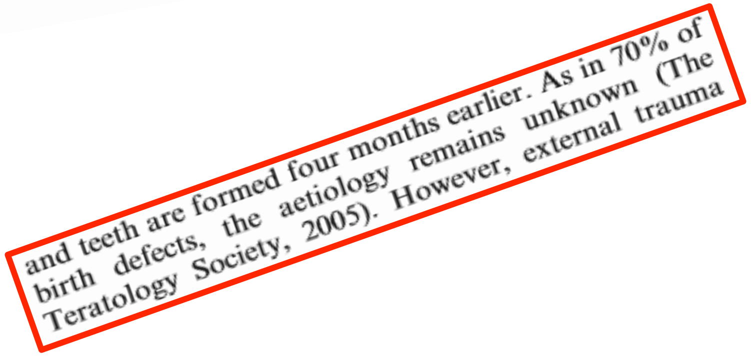
Reprod Dom Anim **51**, 330–334 (2016); doi: 10.1111/rda.12665
ISSN 0936-6768

Short Communication

***Campylorrhinus lateralis*, Bilateral *microphthalmia* and *odontoma temporalis* in an Oldenburg Foal**

C Casteleyn[1], P Cornillie[2], V Tüllmann[2], S Van Cruchten[1] and C Van Ginneken[1]

[1]*Laboratory of Applied Veterinary Morphology, Department of Veterinary Sciences, Faculty of Pharmaceutical, Biomedical and Veterinary Sciences, University of Antwerp, Wilrijk, Belgium;* [2]*Department of Morphology, Faculty of Veterinary Medicine, Ghent University, Merelbeke, Belgium*


and teeth are formed four months earlier. As in 70% of birth defects, the aetiology remains unknown (The Teratology Society, 2005). However, external trauma

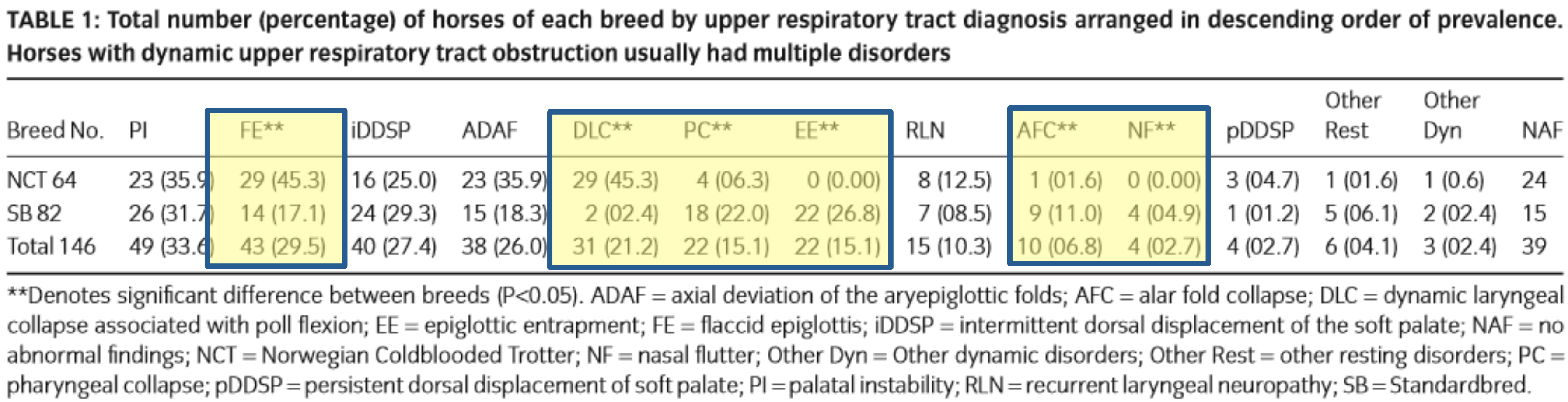


TABLE 1: Total number (percentage) of horses of each breed by upper respiratory tract diagnosis arranged in descending order of prevalence. Horses with dynamic upper respiratory tract obstruction usually had multiple disorders

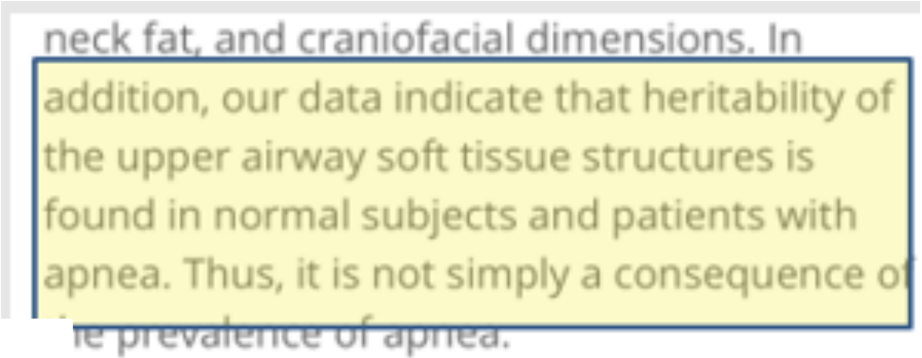
Breed No.	PI	FE**	iDDSP	ADAF	DLC**	PC**	EE**	RLN	AFC**	NF**	pDDSP	Other Rest	Other Dyn	NAF
NCT 64	23 (35.9)	29 (45.3)	16 (25.0)	23 (35.9)	29 (45.3)	4 (06.3)	0 (0.00)	8 (12.5)	1 (01.6)	0 (0.00)	3 (04.7)	1 (01.6)	1 (0.6)	24
SB 82	26 (31.7)	14 (17.1)	24 (29.3)	15 (18.3)	2 (02.4)	18 (22.0)	22 (26.8)	7 (08.5)	9 (11.0)	4 (04.9)	1 (01.2)	5 (06.1)	2 (02.4)	15
Total 146	49 (33.6)	43 (29.5)	40 (27.4)	38 (26.0)	31 (21.2)	22 (15.1)	22 (15.1)	15 (10.3)	10 (06.8)	4 (02.7)	4 (02.7)	6 (04.1)	3 (02.4)	39

**Denotes significant difference between breeds (P<0.05). ADAF = axial deviation of the aryepiglottic folds; AFC = alar fold collapse; DLC = dynamic laryngeal collapse associated with poll flexion; EE = epiglottic entrapment; FE = flaccid epiglottis; iDDSP = intermittent dorsal displacement of the soft palate; NAF = no abnormal findings; NCT = Norwegian Coldblooded Trotter; NF = nasal flutter; Other Dyn = Other dynamic disorders; Other Rest = other resting disorders; PC = pharyngeal collapse; pDDSP = persistent dorsal displacement of soft palate; PI = palatal instability; RLN = recurrent laryngeal neuropathy; SB = Standardbred.

Heritability of upper airway soft tissue structures

structural vs. dynamic?

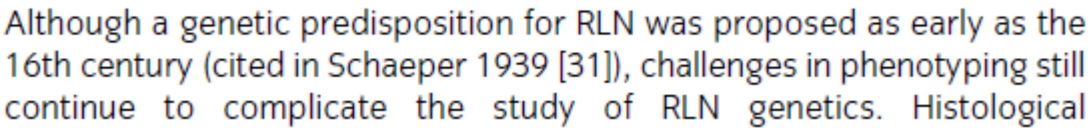
normal vs. diseased?



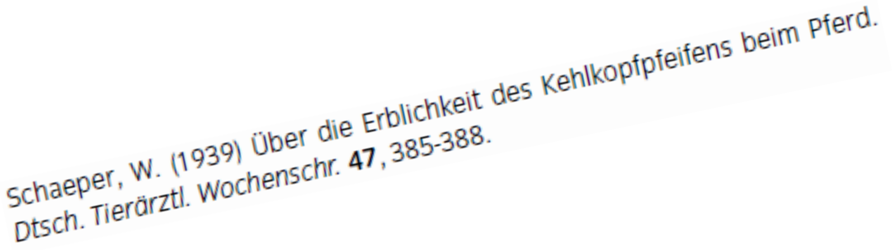
neck fat, and craniofacial dimensions. In addition, our data indicate that heritability of the upper airway soft tissue structures is found in normal subjects and patients with apnea. Thus, it is not simply a consequence of

he prevalence of apnea.

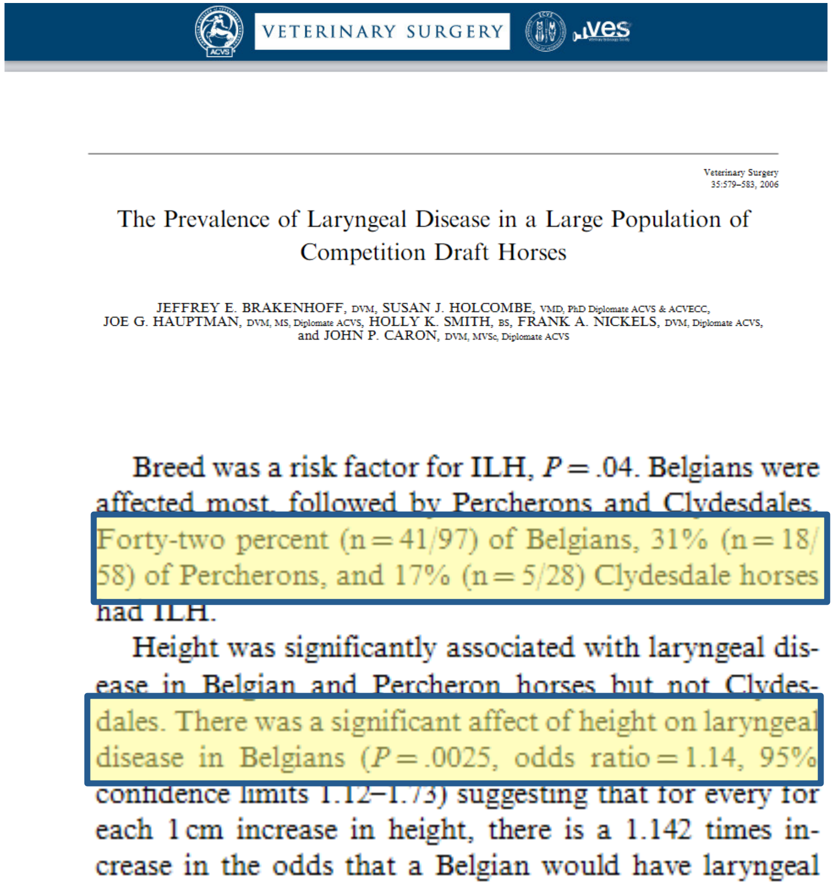
RLN – breed differences



Although a genetic predisposition for RLN was proposed as early as the 16th century (cited in Schaeper 1939 [31]), challenges in phenotyping still continue to complicate the study of RLN genetics. Histological



Schaeper, W. (1939) Über die Erblichkeit des Kehlkopfpfeifens beim Pferd. Dtsch. Tierärztl. Wochenschr. **47**, 385-388.



VETERINARY SURGERY

Veterinary Surgery
35:579–583, 2006

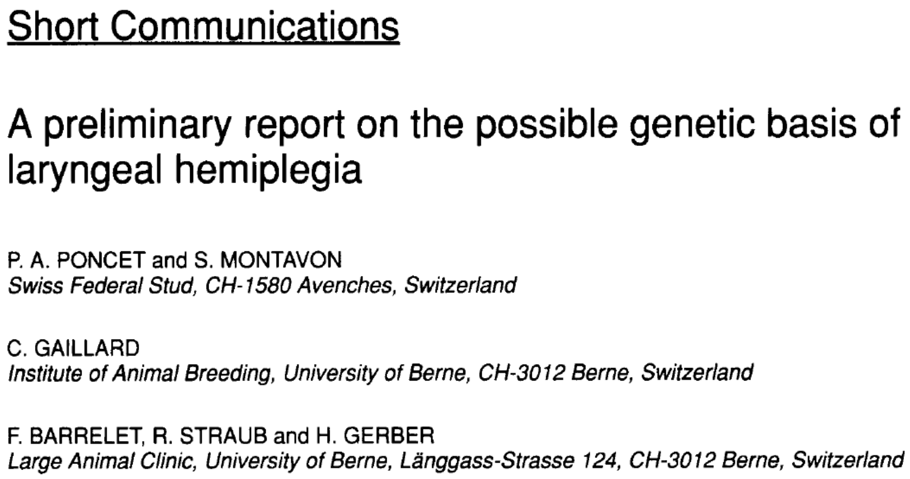
The Prevalence of Laryngeal Disease in a Large Population of Competition Draft Horses

JEFFREY E. BRAKENHOFF, DVM, SUSAN J. HOLCOMBE, VMD, PhD Diplomate ACVS & ACVECC,
JOE G. HAUPTMAN, DVM, MS, Diplomate ACVS, HOLLY K. SMITH, BS, FRANK A. NICKELS, DVM, Diplomate ACVS,
and JOHN P. CARON, DVM, MVSc, Diplomate ACVS

Breed was a risk factor for ILH, $P = .04$. Belgians were affected most, followed by Percherons and Clydesdales. Forty-two percent (n = 41/97) of Belgians, 31% (n = 18/58) of Percherons, and 17% (n = 5/28) Clydesdale horses had ILH.

Height was significantly associated with laryngeal disease in Belgian and Percheron horses but not Clydesdales. There was a significant affect of height on laryngeal disease in Belgians ($P = .0025$, odds ratio = 1.14, 95% confidence limits 1.12–1.73) suggesting that for every for each 1 cm increase in height, there is a 1.142 times increase in the odds that a Belgian would have laryngeal

RLN – halfsib families



Short Communications

A preliminary report on the possible genetic basis of laryngeal hemiplegia

P. A. PONCET and S. MONTAVON
Swiss Federal Stud, CH-1580 Avenches, Switzerland

C. GAILLARD
Institute of Animal Breeding, University of Berne, CH-3012 Berne, Switzerland

F. BARRELET, R. STRAUB and H. GERBER
Large Animal Clinic, University of Berne, Länggass-Strasse 124, CH-3012 Berne, Switzerland

47 % vs. 10 %

Boyko *et al. BMC Genomics* 2014, **15**:259
http://www.biomedcentral.com/1471-2164/15/259

BMC Genomics

RESEARCH ARTICLE **Open Access**

Genomic analysis establishes correlation between growth and laryngeal neuropathy in Thoroughbreds

Adam R Boyko[1], Samantha A Brooks[2], Ashley Behan-Braman[3], Marta Castelhano[4], Elizabeth Corey[4], Kyle C Oliveira[1], June E Swinburne[5,6], Rory J Todhunter[4], Zhiwu Zhang[4], Dorothy M Ainsworth[4] and Norman Edward Robinson[3*]

Abstract

Background: Equine recurrent laryngeal neuropathy (RLN) is a bilateral mononeuropathy with an unknown pathogenesis that significantly affects performance in Thoroughbreds. A genetic contribution to the pathogenesis of RLN is suggested by the higher prevalence of the condition in offspring of RLN-affected than unaffected stallions. To better understand RLN pathogenesis and its genetic basis, we performed a genome-wide association (GWAS) of 282 RLN-affected and 268 control Thoroughbreds.

Results: We found a significant association of RLN with the *LCORL/NCAPG* locus on ECA3 previously shown to affect body size in horses. Using height at the withers of 505 of these horses, we confirmed the strong association of this locus with body size, and demonstrated a significant phenotypic and genetic correlation between height and RLN grade in this cohort. Secondary genetic associations for RLN on ECA18 and X did not correlate with withers height in our cohort, but did contain candidate genes likely influencing muscle physiology and growth: myostatin (*MSTN*) and integral membrane protein 2A (*ITM2A*).

Conclusions: This linkage between body size and RLN suggests that selective breeding to reduce RLN prevalence would likely reduce adult size in this population. However, our results do not preclude the possibility of modifier loci that attenuate RLN risk without reducing size or performance, or that the RLN risk allele is distinct but tightly linked to the body size locus on ECA3. This study is both the largest body size GWAS and the largest RLN GWAS within Thoroughbred horses to date, and suggests that improved understanding of the relationship between genetics, equine growth rate, and RLN prevalence may significantly advance our understanding and management of this disease.

Keywords: Recurrent laryngeal neuropathy (RLN), Thoroughbred, Horse, *Equus caballus*, Genome-wide association study (GWAS), Haplotype, Body size

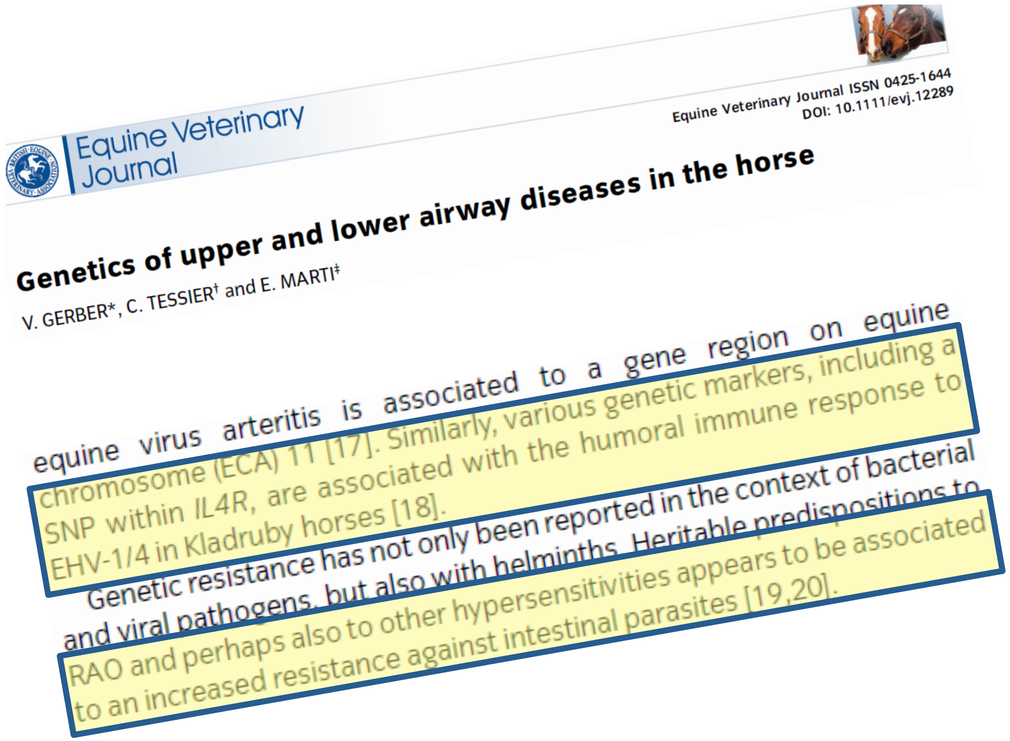
Genomics & Thoroughbred industry EFTBA and ITBF Autumn 2015

RLN moderate to high heritability
Different chromosomal regions
«Science – commercial vs. Industry interests»

EFTBA & ITBF –

- Refused permission for retrospective access to DNA stores held by Pedigree registration authorities by these companies, which could have rapid far reaching consequences, and has insisted that the application of genomics should be gradual and measured.
- Stated that they are against any linkage of DNA performance profiles / genomics to sales catalogues
- Cautiously accepted that there may be a value in the investigation of disease traits, and has created and sought ongoing <u>independent</u> advice from the Equine Genomics Advisory Group*.

Infectious diseases?


Equine Veterinary Journal

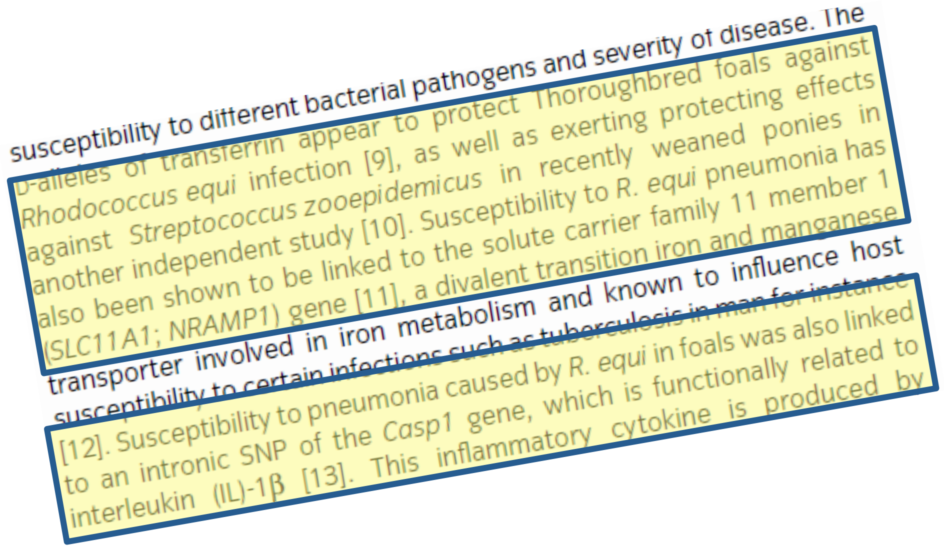
Equine Veterinary Journal ISSN 0425-1644
DOI: 10.1111/evj.12289

Genetics of upper and lower airway diseases in the horse

V. GERBER*, C. TESSIER† and E. MARTI‡

equine virus arteritis is associated to a gene region on equine chromosome (ECA) 11 [17]. Similarly, various genetic markers, including a SNP within *IL4R*, are associated with the humoral immune response to EHV-1/4 in Kladruby horses [18].

Genetic resistance has not only been reported in the context of bacterial and viral pathogens, but also with helminths. Heritable predispositions to RAO and perhaps also to other hypersensitivities appears to be associated to an increased resistance against intestinal parasites [19,20].


susceptibility to different bacterial pathogens and severity of disease. The D-alleles of transferrin appear to protect Thoroughbred foals against *Rhodococcus equi* infection [9], as well as exerting protecting effects against *Streptococcus zooepidemicus* in recently weaned ponies in another independent study [10]. Susceptibility to *R. equi* pneumonia has also been shown to be linked to the solute carrier family 11 member 1 (*SLC11A1; NRAMP1*) gene [11], a divalent transition iron and manganese transporter involved in iron metabolism and known to influence host susceptibility to certain infections such as tuberculosis in man for instance [12]. Susceptibility to pneumonia caused by *R. equi* in foals was also linked to an intronic SNP of the *Casp1* gene, which is functionally related to interleukin (IL)-1β [13]. This inflammatory cytokine is produced by

EIPH – is it hereditary, or not?

Contents lists available at ScienceDirect

The Veterinary Journal

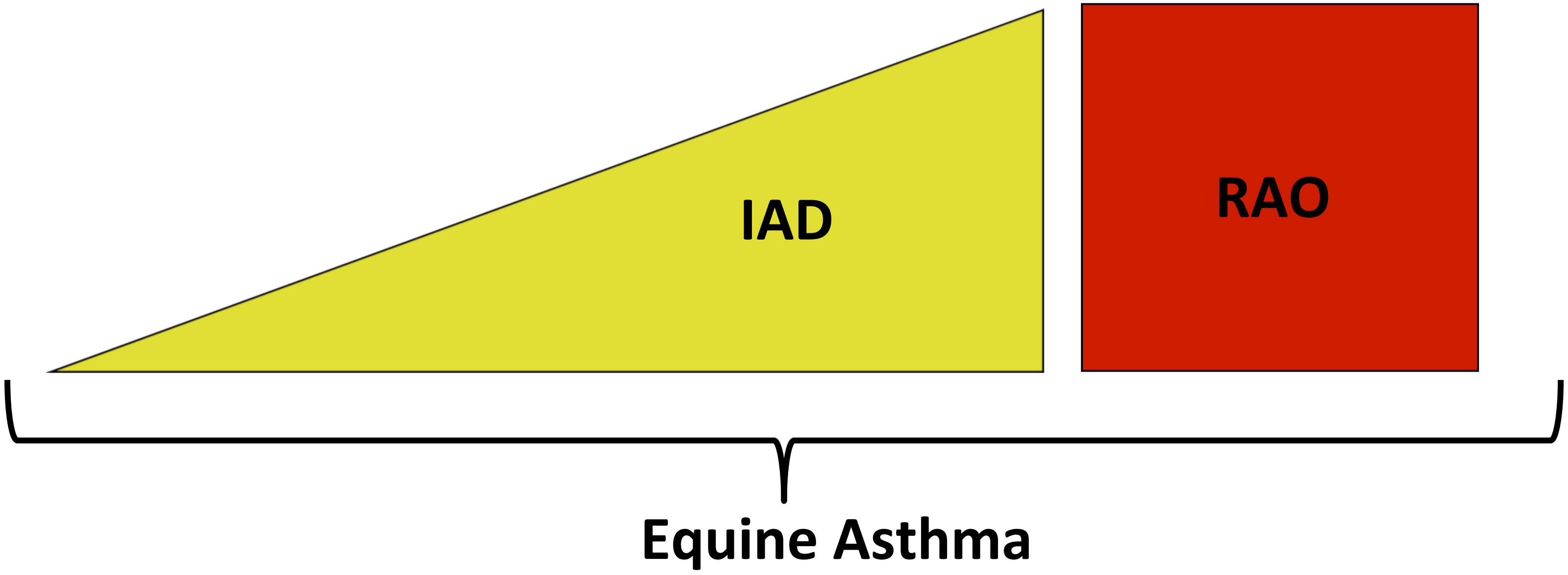
journal homepage: www.elsevier.com/locate/tvjl

Heritability of epistaxis in the Australian Thoroughbred racehorse population

B.D. Velie [a,*], H.W. Raadsma [b], C.M. Wade [a], P.K. Knight [c,1], N.A. Hamilton [a,1]

[a] *Faculty of Veterinary Science, University of Sydney, Camperdown, Sydney, New South Wales 2006, Australia*
[b] *ReproGen-Animal Bioscience Group, Faculty of Veterinary Science, University of Sydney, Camden, New South Wales 2570, Australia*
[c] *Discipline of Biomedical Science, Sydney Medical School, Lidcombe, New South Wales 2141, Australia*

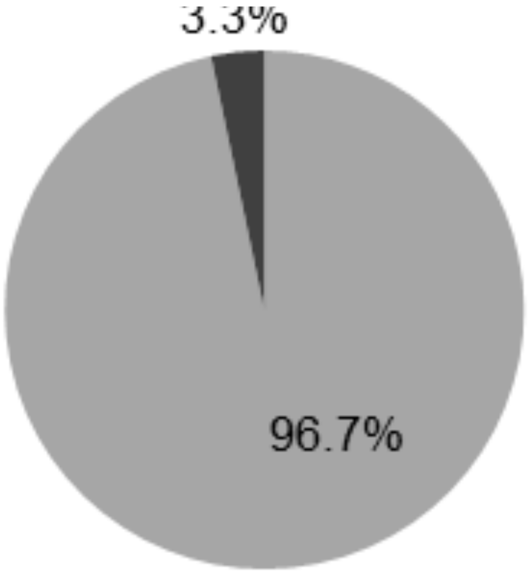
Equine asthma – genetic effects? It depends



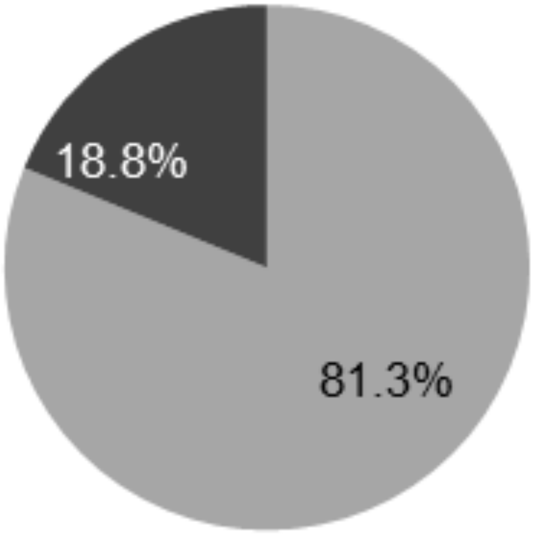
J Vet Intern Med 2014;28:618–623

Evaluation of Coughing and Nasal Discharge as Early Indicators for An Increased Risk to Develop Equine Recurrent Airway Obstruction (RAO)

S. Bosshard and V. Gerber



no clinical signs



occasional cough

Horses with occasional cough +/- nasal discharge have a > 6-10 x increased risk to develop RAO / severe asthma within 2-3 years

Bosshard et al., 2014

Schäper, 1939

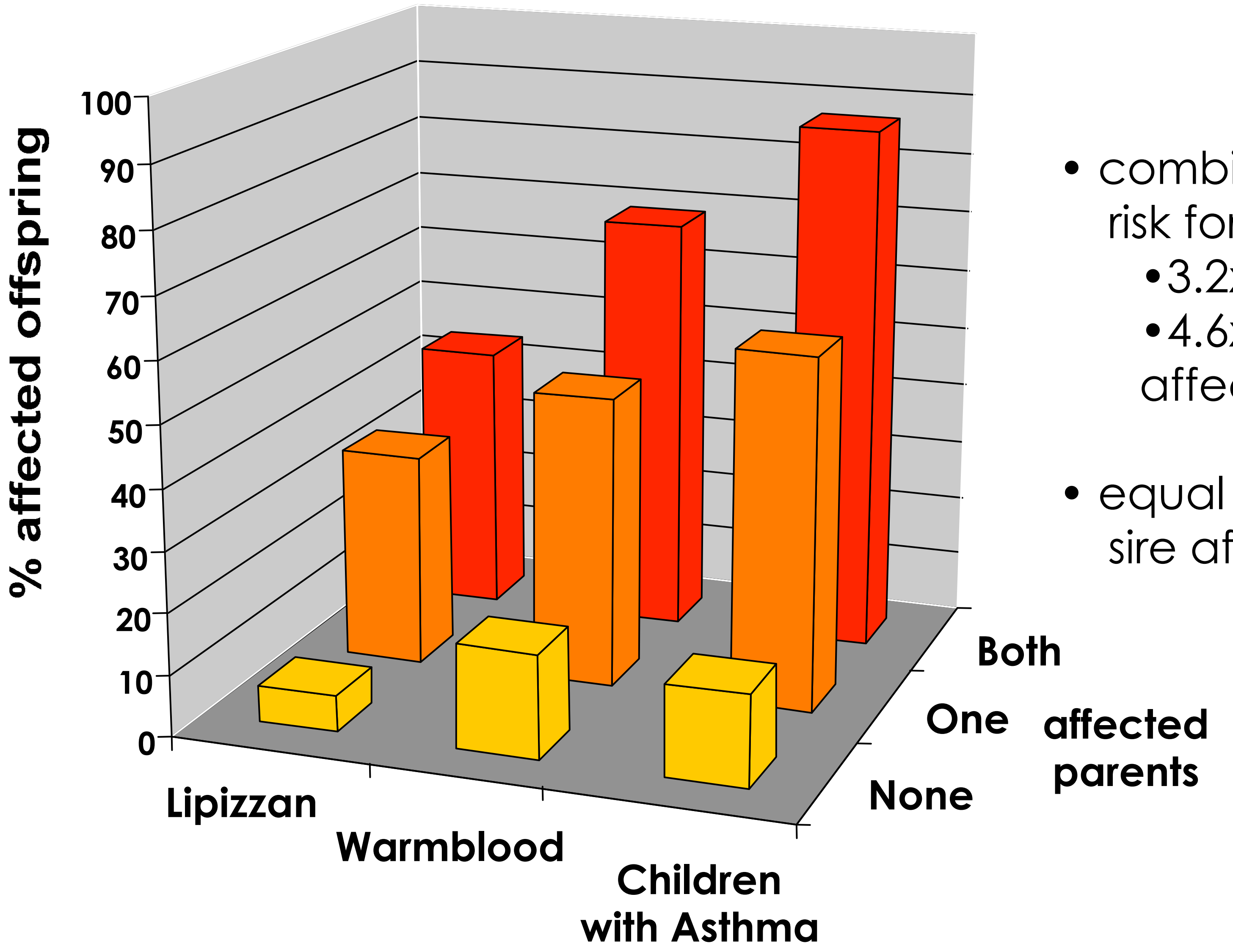
Koch, 1957

Gerber, 1989

Marti et al., 1991

increased risk of RAO
in families of different breeds
suggesting a genetic basis

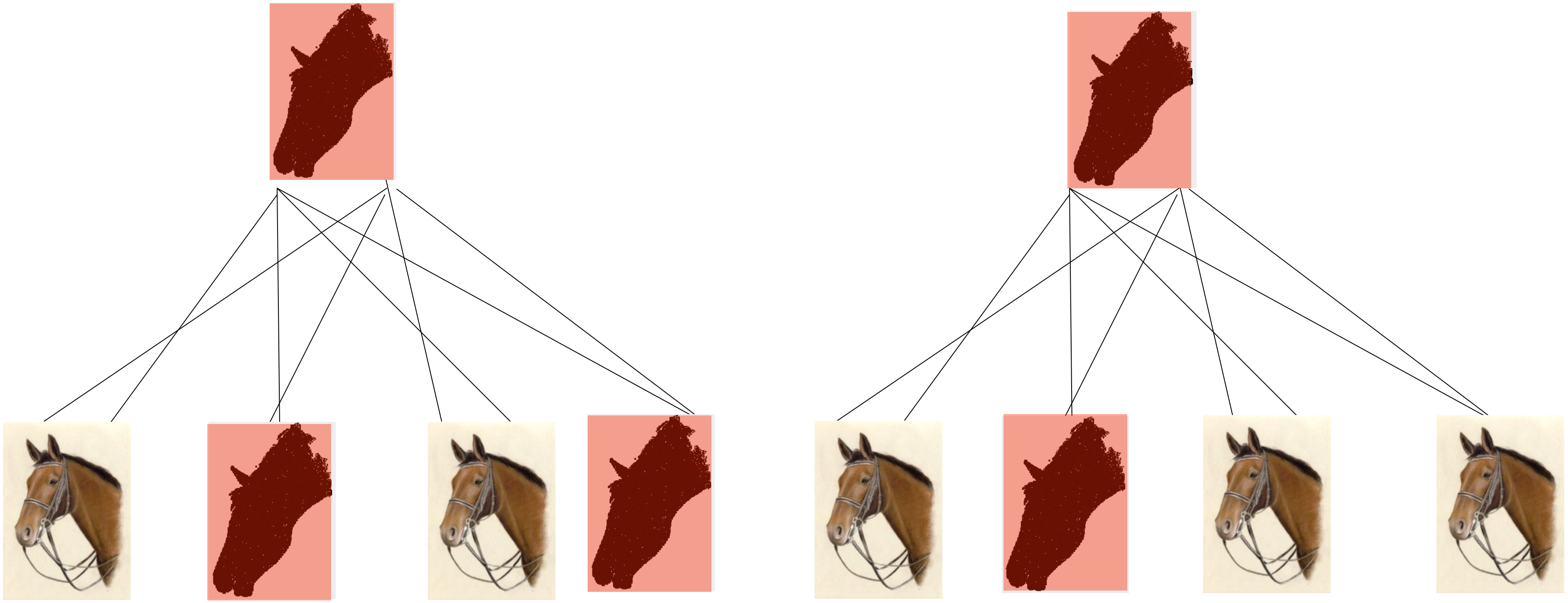
RAO and Asthma – effect of parents



- combined relative risk for RAO:
  - 3.2x with one
  - 4.6x with two affected parents
- equal risk - dam or sire affected

Marti et al. 1991

Investigation of two half-sibling Families of Warmblood Horses



RAO-affected
sire 1

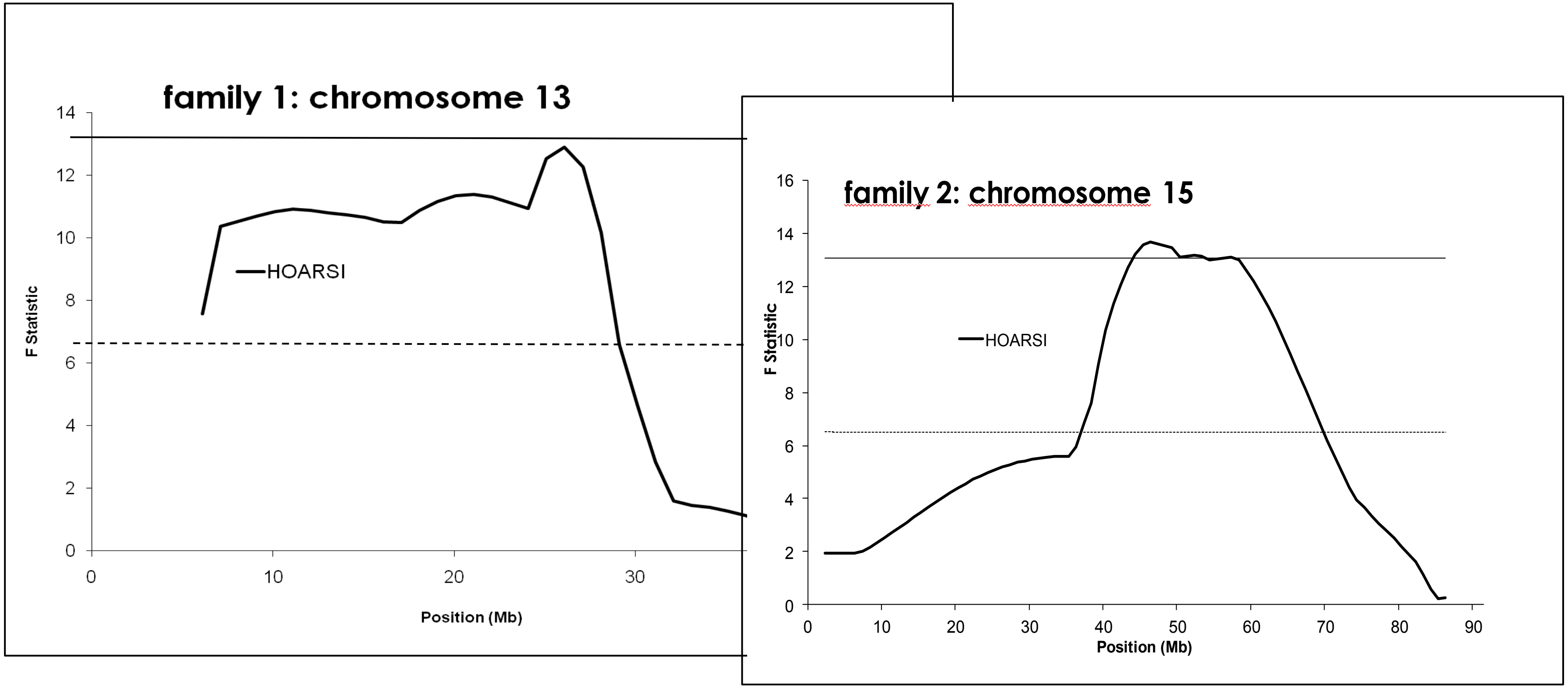
RAO-affected
sire 2

Both sires have >200 offspring aged 5 -15 years

Whole–Genome–Scan

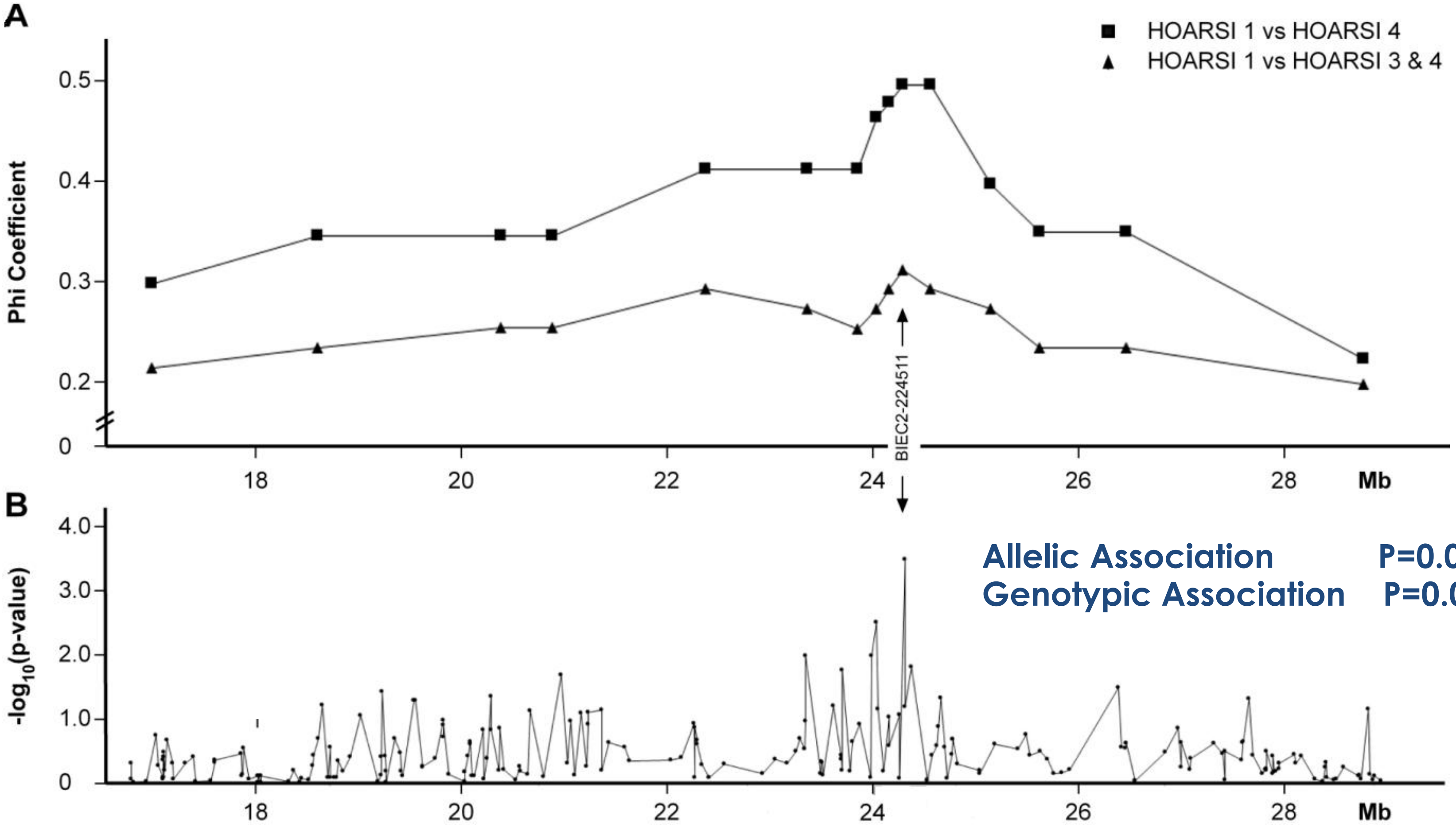
252 microsatellite markers across the 31 autosomes

Identified 2 chromosomal regions associated with RAO



Swinburne et al., 2009 Mamm Genome

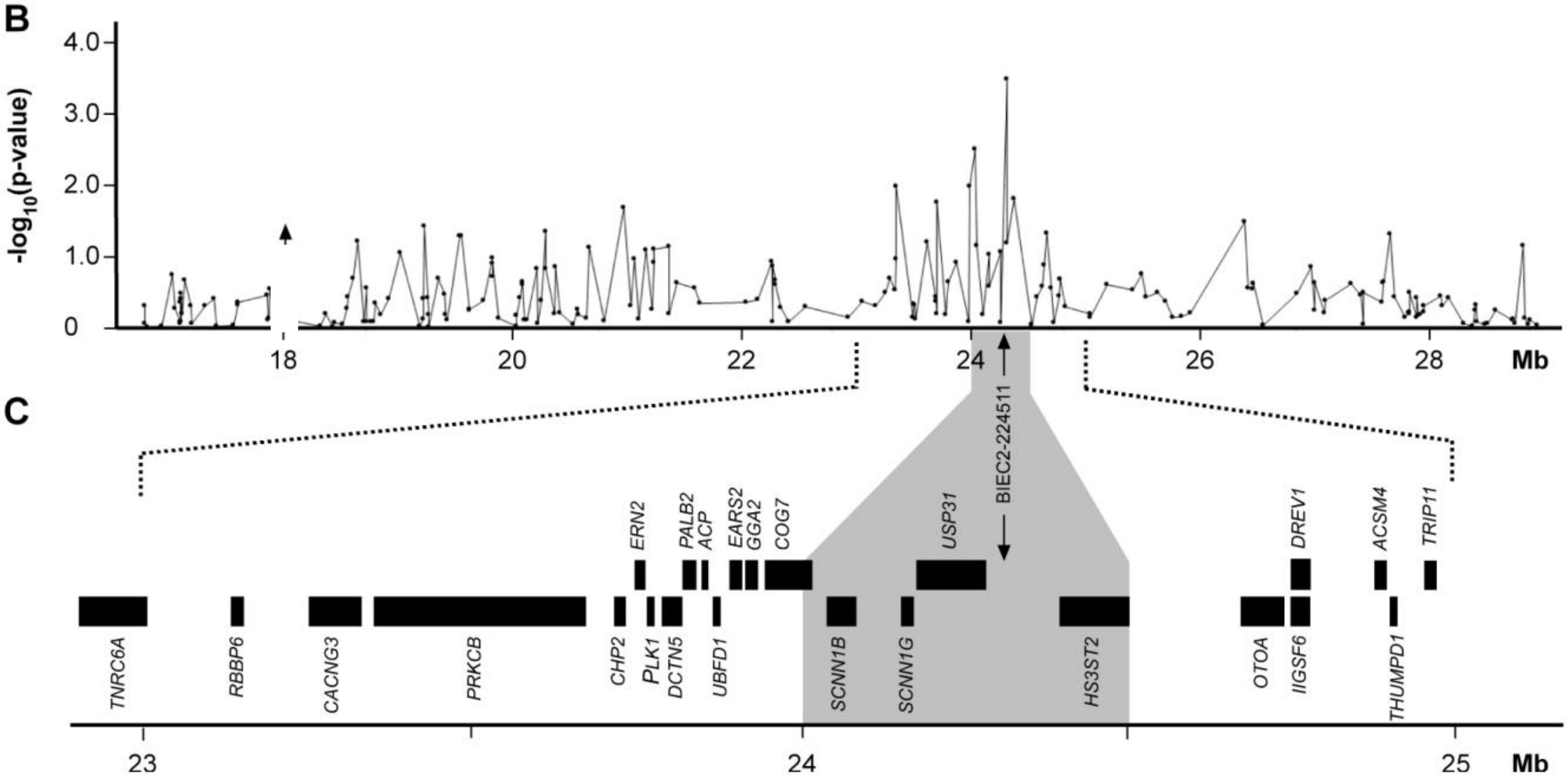
Replication in 646 unrelated Horses



Allelic Association **P=0.004664**
Genotypic Association **P=0.00037**

Shaksi-Niaei et al. 2011, Animal Genetics

Positional Candidate Genes



Shaksi-Niaei et al. 2011, Animal Genetics

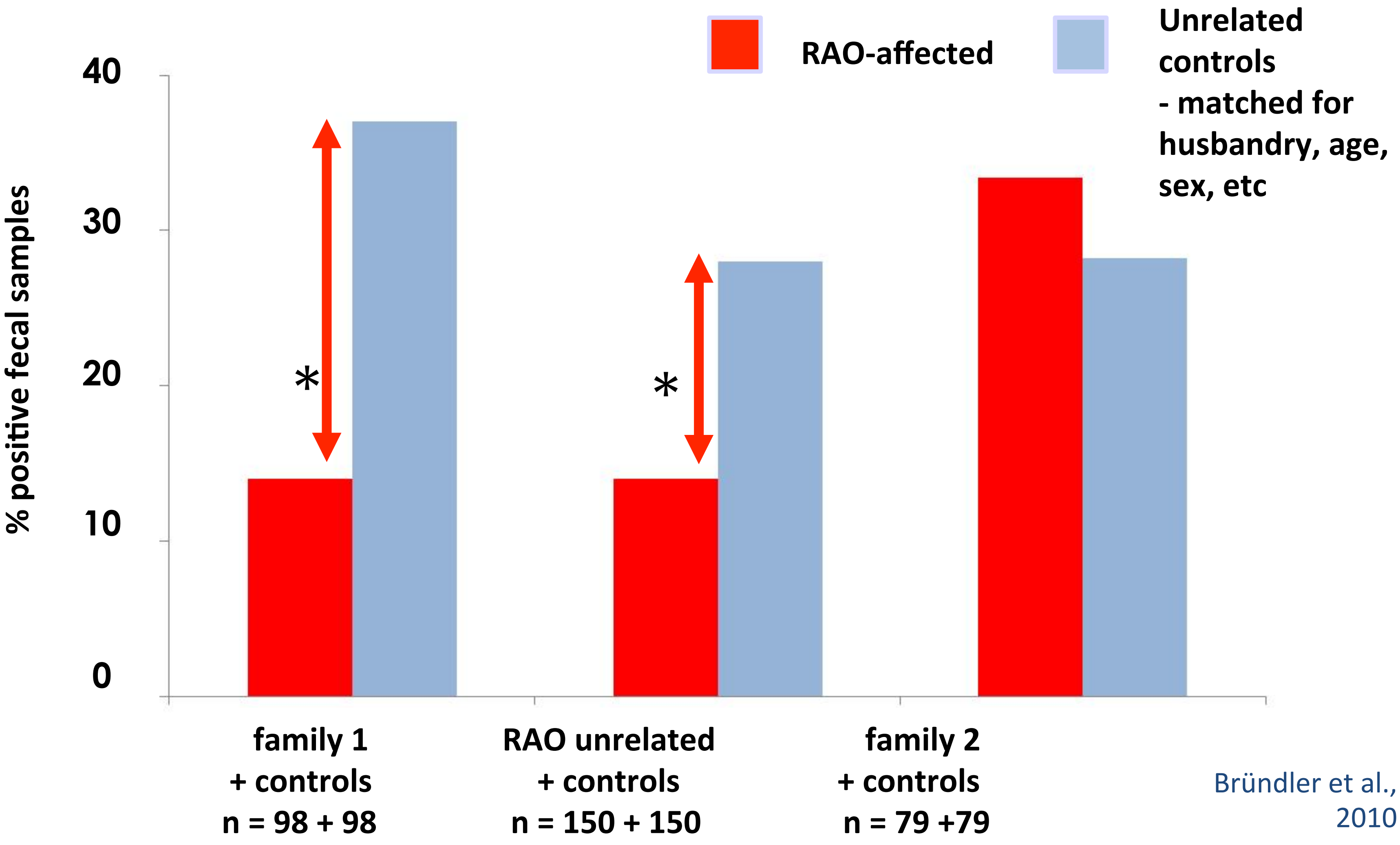
Il4Ralpha: Interesting Candidate Gene

family 1: strong genetic association
and increased gene expression

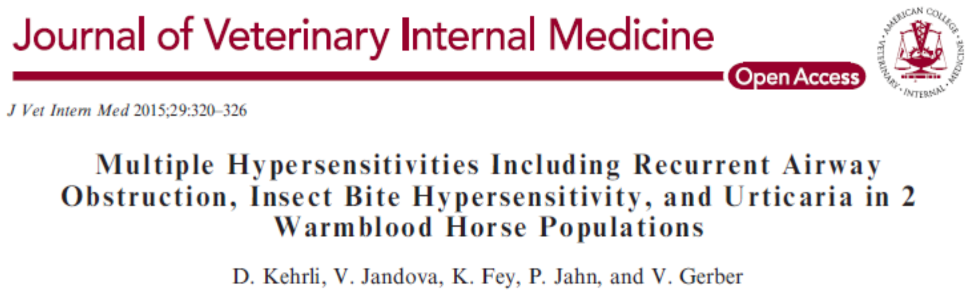
Klukowska et al. 2006, Anim Genet
Jost et al. 2007, Eq Vet J
Klukowska et al. 2012

Humans and other species:
Associated with skin allergies and with parasite defense

RAO and Strongylid Egg Shedding



Bründler et al., 2010



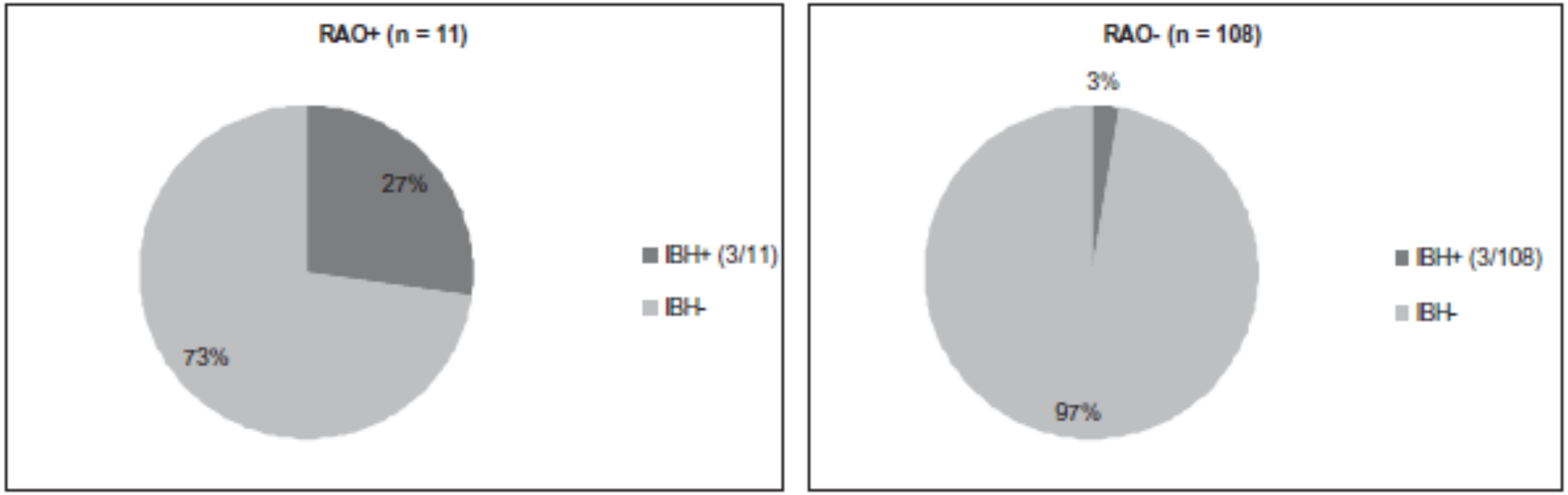
Journal of Veterinary Internal Medicine
Open Access

J Vet Intern Med 2015;29:320–326

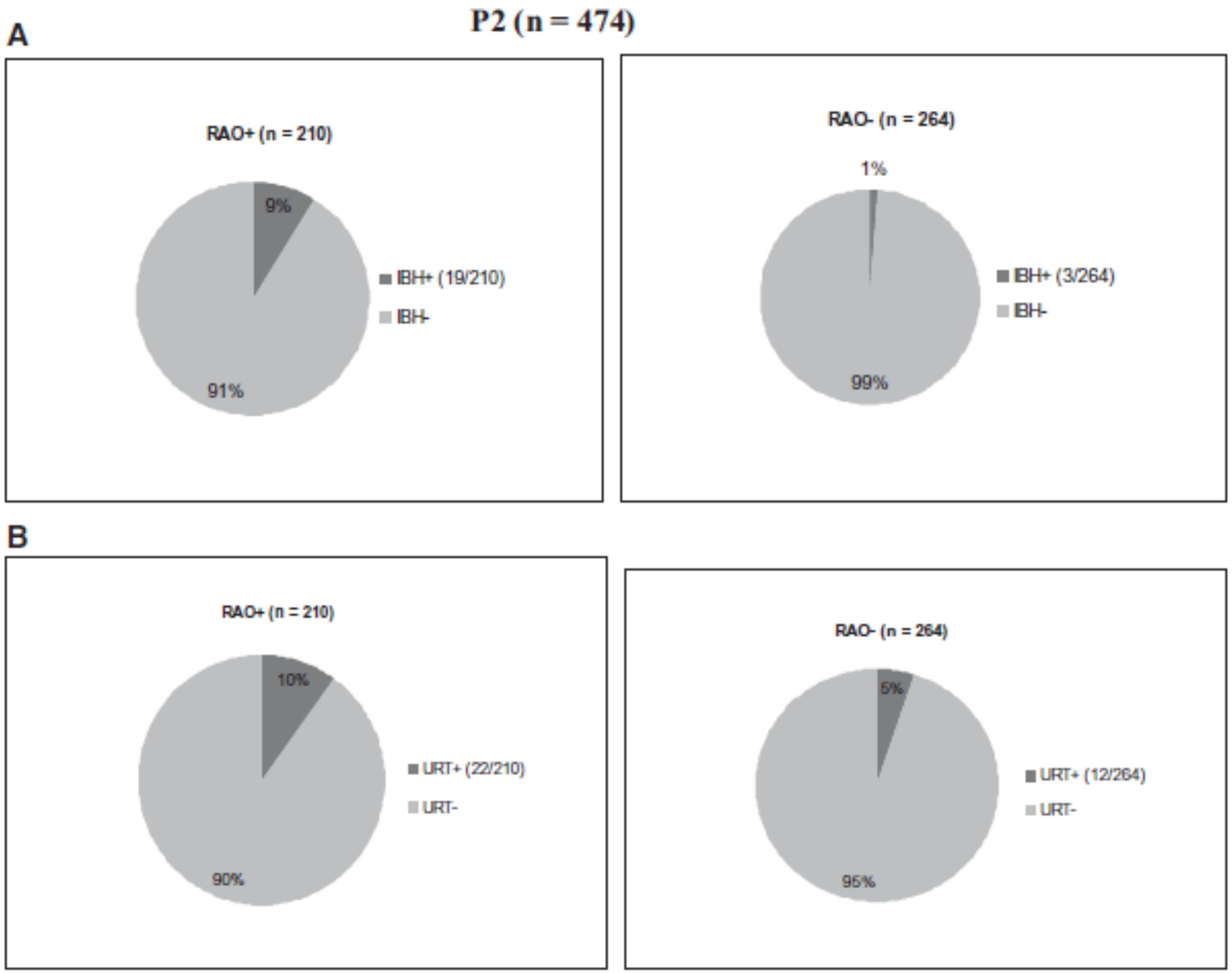
Multiple Hypersensitivities Including Recurrent Airway Obstruction, Insect Bite Hypersensitivity, and Urticaria in 2 Warmblood Horse Populations

D. Kehrli, V. Jandova, K. Fey, P. Jahn, and V. Gerber

P1 (n = 119)



In P1, RAO-affected horses had a 13.1 times higher odds ratio (OR) of also suffering from IBH

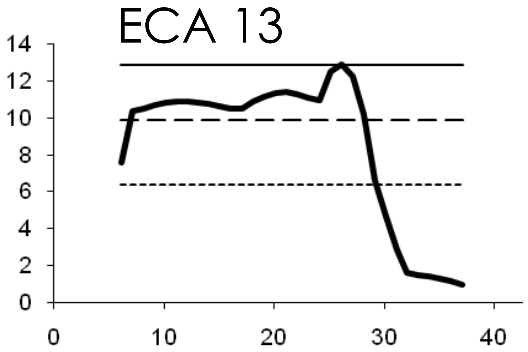


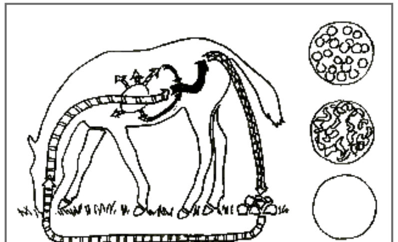
In P2:

- RAO - affected horses showed a 7.4 times higher OR of concomitantly suffering from IHB
- RAO - affected horses showed a 7.1 times higher OR of concomitantly suffering from URT

Family 1

rezessive
mode of
inheritance





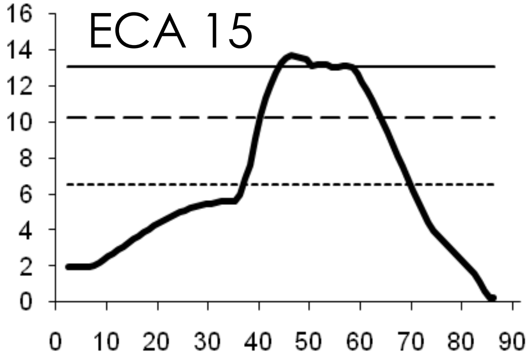
resistent

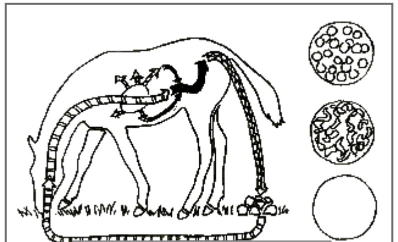
?

same RAO phenotype

Family 2

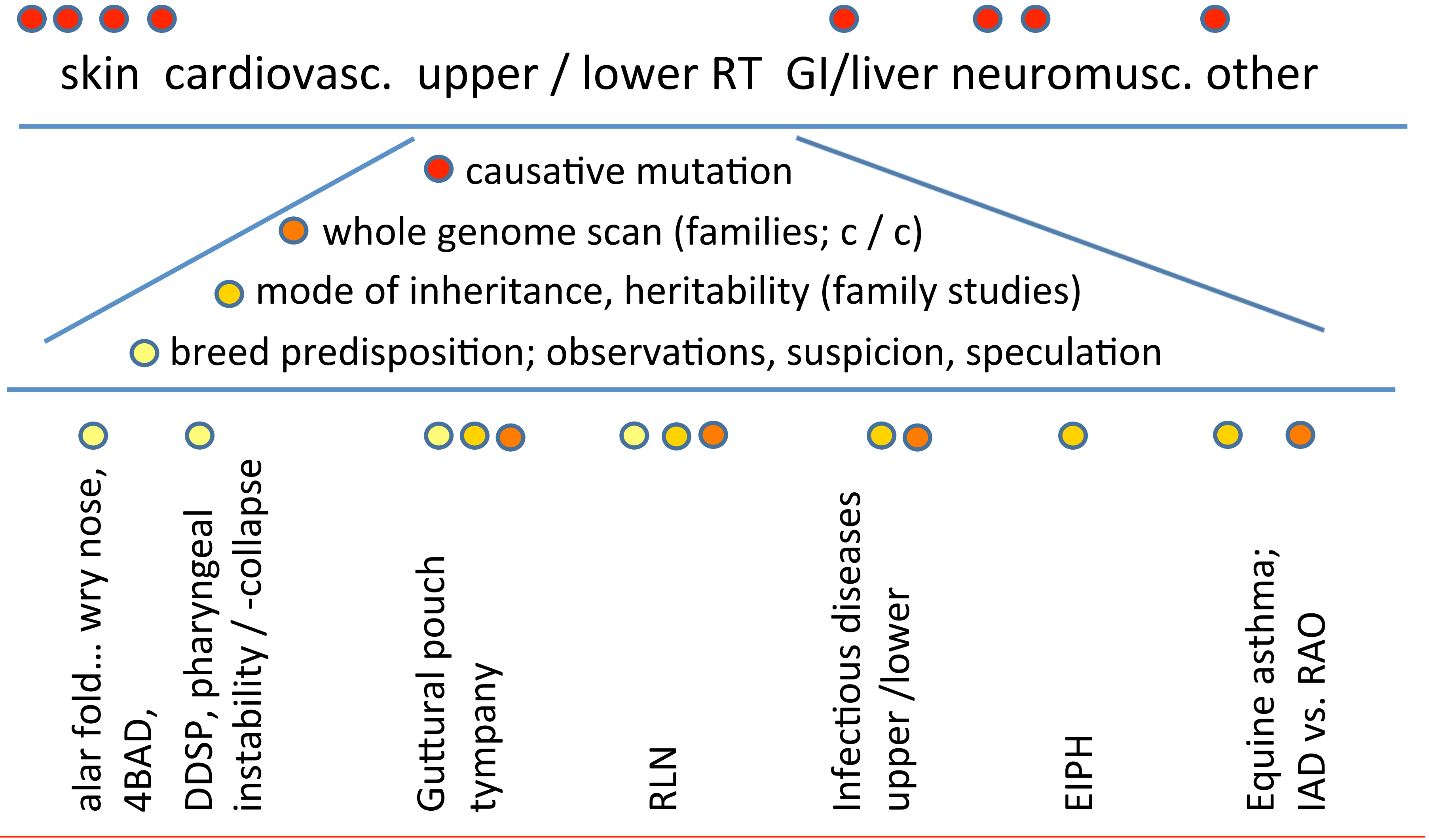
dominant
mode of
inheritance





normal

Medical genetics of the horse by organ system & disorder



skin cardiovasc. upper / lower RT GI/liver neuromusc. other

● causative mutation

● whole genome scan (families; c / c)

● mode of inheritance, heritability (family studies)

● breed predisposition; observations, suspicion, speculation

alar fold… wry nose, 4BAD,

DDSP, pharyngeal instability / -collapse

Guttural pouch tympany

RLN

Infectious diseases upper /lower

EIPH

Equine asthma; IAD vs. RAO

Collaborations and Acknowledgments

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