

# **Antigen array for serological diagnosis and novel allergen identification in severe equine asthma**

S. White<sup>1, 2, 3\*</sup>, M. Moore-Colyer<sup>1</sup>, E. Marti<sup>4</sup>, D. Hannant<sup>5</sup>, V. Gerber<sup>4</sup>, L. Coüetil<sup>6</sup>, E.A. Richard<sup>7,</sup>  
<sup>8</sup> and M. Alcocer<sup>2</sup>.

<sup>1</sup>Royal Agricultural University, Cirencester, Gloucestershire, GL7 6JS, UK.

<sup>2</sup>School of Biosciences, University of Nottingham, Sutton Bonington Campus, Loughborough, LE12 5RD, UK.

<sup>3</sup>Nottingham Trent University, Brackenhurst Campus, Southwell, Nottinghamshire, NG25 0QF, UK.

<sup>4</sup>Department of Clinical Research and Veterinary Public Health, University of Bern, Bremgartenstr, Postfach, 3001 Bern, Switzerland.

<sup>5</sup>School of Veterinary Medicine and Science, University of Nottingham, Sutton Bonington Campus, Loughborough, LE12 5RD, UK.

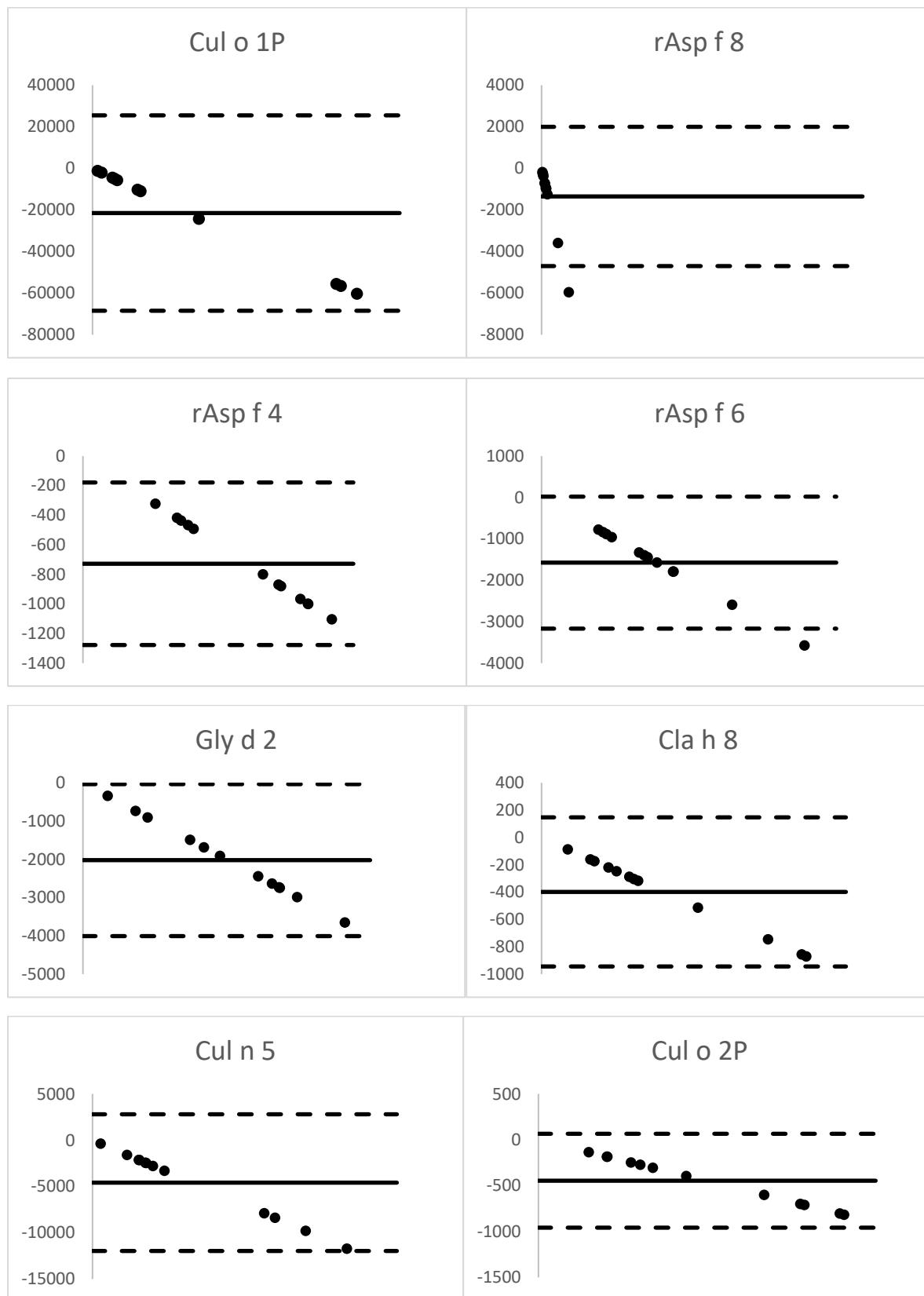
<sup>6</sup>Veterinary Clinical Sciences, College of Veterinary Medicine, Purdue University, West Lafayette, IN, 47907, USA.

<sup>7</sup>LABÉO Frank Duncombe, 1 route de Rosel, 14053 Caen Cedex 4, France.

<sup>8</sup>Normandie Univ, UniCaen, BIOTARGEN, 3 rue Nelson Mandela, 14280 Saint-Contest, France.

\*samuel.white@ntu.ac.uk

Bland Altman plots to compare ELISA and microarray IgE with specific proteins. The acceptable level of bias is dictated by the solid line. The dashed lines represent the 95% limits of agreement as the mean difference (2 SD).



Summary of horses used in the study.

	<b>Switzerland</b>	<b>France</b>	<b>USA</b>	<b>Canada</b>
Number of horses	16 sEA affected 24 IBH affected 23 IBH/sEA affected 40 control	5 sEA affected 6 control	6 sEA affected 6 control	6 sEA affected 6 control
Horse Owner Assessed Respiratory Signs Index (HOARSI) (Ramseyer et al., 2007)	†	na	na	na
Histamine/Metacholin Bronchoprovocation : (concentration of histamine when inhalation was stopped)	†	na	na	na
Partial pressure of arterial oxygen	†	na	na	na
Provocation concentration	†	na	na	na
Pulmonary function test	†	†	†	†
Insect bite hypersensitivity score (Etude, 2014)	†	na	na	na
Physical examination : including respiratory and cardiac auscultation, rectal temperature, capillary refill time and colour of mucous membranes, palpation of submandibular lymph nodes, lung auscultation at rest and during rebreathing	†	†	†	†
Mucus scoring system	na	†	†	†
Bronchoalveolar lavage fluid (including cytology)	na	†	†	†
Owner reported coughing, nasal discharge at time of examination	†	†	†	†
Reversible airway obstruction (after medical/environmental change)	na	†	na	na

† : measured in this group

na : not analysed

Variable influences on the projection scores significant for class prediction from the environmentally mixed group of horses (n=138) after VIP selection.

<b>Species</b>	<b>Allergome name</b>	<b>Extract = E; Molecule = M</b>	<b>VIP score</b>
<i>Hevea brasiliensis</i>	Hev b 11	M	2.397886012
<i>Hevea brasiliensis</i>	Hev b 6.02	M	2.242175574
<i>Hevea brasiliensis</i>	Hev b 5.0101	M	2.061708356
<i>Aspergillus fumigatus</i>	rAsp f 8	M	2.034462948
<i>Helix aspersa</i>	Hel as 7	M	1.693216892
<i>Culicoides obsoletus</i>	Cul o 145	M	1.553348148
<i>Hevea brasiliensis</i>	Hev b 3.0101	M	1.447433432
<i>Betula verrucosa</i>	Bet v 2.0101	M	1.439197666
<i>Culicoides obsoletus</i>	Cul nu 2	M	1.401734766
<i>Culicoides nubeculosus</i>	Cul o1P	M	1.370674276
<i>Blattella germanica</i>	Bla g 2	M	1.294109254
<i>Culicoides obsoletus</i>	Cul o 2	M	1.289809746
<i>Mercurialis annua</i>	Mer a 1	M	1.257180767
<i>Bos domesticus</i>	Bos d LF	M	1.234823184
<i>Culicoides obsoletus</i>	Cul ob 8	M	1.132147319
<i>Prunus persica</i>	Pru p 3	M	1.131899686
<i>Felis domesticus</i>	Fel d [Epithelia]	E	1.131671083
<i>Mucor circinelloides</i>	Muc ci	E	1.113604072
<i>Culicoides obsoletus</i>	C0120	M	1.107017518
<i>Quercus robur</i>	Que r [Pollen]	E	1.103588161
<i>Culicoides obsoletus</i>	Cul ob 8	M	1.10169779
<i>Epicoccum nigrum</i>	Epi p	E	1.097080342
<i>Gallus domesticus</i>	Gal d [Egg White]	E	1.092020409
<i>Thermoactinomyces vulgaris</i>	The v	E	1.078503603
<i>Culicoides obsoletus</i>	Cul n 10.03	M	1.070733631
<i>Hevea brasiliensis</i>	Hev b	E	1.053235133
<i>Ulmus americana</i>	Ulm a	E	1.046124579
<i>Eupatorium capillifolium</i>	Eup c	E	1.041304179
<i>Actinidia deliciosa</i>	Act d 2	M	1.039211209
<i>Glycine max</i>	Soybean AL3-recombinant	M	1.036586734
<i>Parietaria judaica</i>	Par j 1	M	1.025947792
<i>Blomia tropicalis</i>	Blo t	E	1.024506004
<i>Rattus norvegicus</i>	Rat n [Epithelium]	E	1.022811408
<i>Corylus americana</i>	Cor am	E	1.00432009
<i>Culicoides nubeculosus</i>	Cul ob 8	M	0.999496997
<i>Lupinus albus</i>	Lup a [Seed]	E	0.990564278
<i>Culicoides obsoletus</i>	Co167	M	0.983835139

Culicoides obsoletus	Cul o 8C11	M	0.98257687
Eurotium amstelodami	Eur s	E	0.981731287
Penaeus monodon	Pen m 1	M	0.977846295
Zea mays	Zea m [Seed]	E	0.975934894
Kineosporia rhizophila	Kin r	E	0.96268091
Olea europaea	Ole e 1	M	0.957886893
Aspegillus flavus	Asp fl	E	0.954692807
Olea europaea	Ole e [Pollen]	E	0.946553558
Dermatophagoides farinae	Der f 1	M	0.943956016
Geotrichum candidum	Geo c	E	0.934810722
Arachis hypogaea	Ara h 6	M	0.931278284
Helix aspersa	Hel as 7	M	0.929469269
Gallus domesticus	Gal d 4	M	0.92158607
Parietaria judaica	Par j	E	0.919641275
Culicoides	cul o 5	M	0.915549349
Helianthus annuus	Hel a 2S Albumin	M	0.915519266
Canis familiaris	Can f [Epithelium]	E	0.914611185
Solanum lycopersicum	Sola l LTP_7kD	M	0.909716836
Helianthus annuus	Hel a [Pollen]	E	0.898832645
Periplaneta americana	Per a 7	M	0.892563113
Corylus avellana	Cor a [Seed]	E	0.877063468
Culicoides obsoletus	Cul o 3	M	0.87297805
Dermatophagoides pteronyssinus	Der p 2	M	0.866653724
Olea europaea	Ole e 2	M	0.859525551
Fagopyrum esculentum	Fag e	E	0.859249296
Culicoides obsoletus	Cul o 6 F10	M	0.856052813
Eucalyptus globulus	Euc g [Pollen]	E	0.854059354
Quercus ilex	Que i	E	0.842572502
Dermatophagoides pteronyssinus	Der p	E	0.836111501
Actinidia chinensis	Act c 10	M	0.829294987
Artemisia vulgaris	Art v 1	M	0.824564544
Black fly	Sim vi	E	0.823110421
Ricinus communis	Ric c [Pollen]	E	0.815160266
Drechslera halodes	Dre h	E	0.807378063
Culicoides nubeculosus	Cul o 1	M	0.799266712
Hevea brasiliensis	Hev b 9	M	0.796595977
Aspergillus versicolor	Asp v	E	0.796427599
Avena sativa	Ave s [Seed]	E	0.790808149
Actinidia chinensis	Act c 11	M	0.787068315
Manioc	Man e	E	0.785608616
Glycine max	Gly m Agglutinin	M	0.78479311
Glycine max	Gly m TI	M	0.781228253

<i>Periplaneta americana</i>	Per a 7 (0.5mg)	M	0.779612014
<i>Mus musculus</i>	Mus m 1	M	0.777665393
<i>Culicoides obsoletus</i>	Cul o 3	M	0.776157103
hay dust	Hay d	E	0.774529898
<i>Ceratonia siliqua</i>	Cer si [Seed]	E	0.767985414
<i>Culicoides nubeculosus</i>	Cul nu 12	M	0.765522432
<i>Blatta orientalis</i>	Bla o	E	0.760381536
<i>Castanea sativa</i>	Cas s [Pollen]	E	0.748034147
<i>Armoracia rusticana</i>	Arm r HRP	E	0.740446527
<i>Penicillium expansum</i>	Pen e	E	0.734560812
<i>Ctenocephalides felis</i>	Cte f	E	0.716428089
<i>Phleum pratense</i>	Phl p 1.0102	M	0.712552232
<i>Hevea brasiliensis</i>	Hev b 8	M	0.710739268
<i>Merluccius hubbsi</i>	Mer hu 1	M	0.71046373
<i>Dermatophagoides pteronyssinus</i>	Der p 10	M	0.7096095
<i>Hevea brasiliensis</i>	Hev b 1	M	0.707560234
<i>Taraxacum officinale</i>	Tar o [Pollen]	E	0.700310884
<i>Acarus siro</i>	Aca s	E	0.698592024
<i>Culicoides</i>	Cul o 6	M	0.686143673
<i>Bos domesticus</i>	Bos d 5.0102	M	0.672595402
<i>Platanus acerifolia</i>	Pla a 8	M	0.669199521
<i>Musa x paradisiaca</i>	Mus xp	E	0.660557
<i>Equus caballus</i>	Equ c [Milk]	E	0.641591098
<i>Cladosporium herbarum</i>	Cla h	E	0.627312615
<i>Triticum aestivum</i>	Tri a [Pollen]	E	0.625247924
<i>Canis familiaris</i>	Can f 1	M	0.61595743
<i>Saccharomyces cerevisiae</i>	Sac c	E	0.600429374
<i>Culicoides obsoletus</i>	Cul nu 2	M	0.598849656
<i>Carpinus betulus</i>	Car b	E	0.571083305
<i>Arachis hypogaea</i>	Ara h 1	M	0.559353341
<i>Alternaria alternata</i>	Alt a 1	M	0.549001691
<i>Leucanthemum vulgare</i>	Leu vu	E	0.505030455
<i>Brassica</i>	Bra n	E	0.496039753
<i>Acer saccharinum</i>	Ace s	E	0.343599366