doi:10.1111/j.1365-2052.2007.01634.x

Mapping of quantitative trait loci controlling tick [*Riphicephalus* (*Boophilus*) *microplus*] resistance on bovine chromosomes 5, 7 and 14

G. Gasparin*, M. Miyata†, L. L. Coutinho‡, M. L. Martinez§,1, R. L. Teodoro§, J. Furlong§, M. A. Machado§, M. V. G. B. Silva§, T. S. Sonstegard¶ and L. C. A. Regitano**

*Departamento de Genética e Evolução, Universidade Federal de São Carlos, Rodovia Washington Luiz km 235, São Carlos/SP 13565-905, Brazil. †Departamento de Análises Clínicas, Universidade Estadual Paulista 'Júlio de Mesquita Filho', Rodovia Araraquara/Jaú km 1, Araraquara/SP 14801-902, Brazil. ‡ESALQ/USP Escola Superior de Agricultura 'Luiz de Queiroz'/Universidade de São Paulo, Avenida Pádua Dias 11, Piracicaba/SP 13418-900, Brazil. §Embrapa Gado de Leite, Rua Eugênio do Nascimento 610, Juiz de Fora/MG 36038-330, Brazil. ¶USDA, ARS, Bovine Functional Genomics Laboratory, Beltsville, MD 20705, USA. **Embrapa Pecuária Sudeste, Rodovia Washington Luiz km 234, São Carlos/SP 13560-970, Brazil

Summary

Differences in domestication and selection processes have contributed to considerable phenotypic and genotypic differences between *Bos taurus* and *Bos indicus* cattle breeds. Of particular interest in tropical and subtropical production environments are those genetic differences between subspecies that underlie the phenotypic extremes in tolerance and susceptibility to parasite infection. In general, *B. taurus* cattle are more susceptible to ectoparasites than *B. indicus* cattle in tropical environments, and much of this difference is under genetic control. To identify genomic regions involved in tick resistance, we developed a *B. taurus* × *B. indicus* F_2 experimental population to map quantitative trait loci (QTL) for resistance to the *Riphicephalus* (*Boophilus*) *microplus* tick. About 300 individuals were measured for parasite load in two seasons (rainy and dry) and genotyped for 23 microsatellite markers covering chromosomes 5, 7 and 14. We mapped a suggestive chromosome-wide QTL for tick load in the rainy season ($P < 0.05$) on chromosome 5. For the dry season, suggestive ($P < 0.10$) chromosome-wide QTL were mapped on chromosomes 7 and 14. The additive effect of the QTL on chromosome 14 corresponds to 3.18% of the total observed phenotypic variance. Our QTL-mapping study has identified different genomic regions controlling tick resistance; these QTL were dependent upon the season in which the ticks were counted, suggesting that the QTL in question may depend on environmental factors.

Keywords cattle, ectoparasites, microsatellite marker, quantitative trait loci, resistance.

Introduction

There are more than a billion cattle in the world, and most of them can be found in tropical or subtropical regions. One of the major constraints of intensifying production in these regions is infection by endoparasites and ectoparasites. Among the ectoparasites, one of the most noxious to cattle is the tick, *Riphicephalus (Boophilus) microplus* (Horak *et al.* 2002). Powell & Reid (1982) describe its preferential geographical distribution to places with high temperature and humidity, like Australia and Brazil. Because Brazil is almost entirely within a tropical climate, the tick has become one of the most prevalent bovine ectoparasites, causing great economic losses in the production system.

There are few global reports on the costs involved in tick control and tick disease treatments. de Castro (1997) estimated that the annual global costs associated with ticks and the diseases that they transmit to cattle amounted to

Address for correspondence

L. C. A. Regitano, Rodovia Washington Luiz, km 234, CP 339, CEP 13560-970, São Carlos/SP, Brazil.
E-mail: luciana@cppse.embrapa.br

[1]In memoriam.

Accepted for publication 7 June 2007

US$13.9 billion to US$18.7 billion. In Brazil, which has the largest commercial cattle herd in the world at 165 million head (ANUALPEC 2005), tick infestations result in estimated annual losses of US$800 million/year (Martinez *et al.* 2006).

Ticks are among the most relevant vectors of diseases affecting livestock, pets and humans because of their ability to host and transmit disease-causing organisms. These include pathogenic protozoa, rickettsia, spirochaetes and viruses (Jongejan & Uilenberg 2004). Only mosquitoes exceed ticks as vectors of disease-causing agents in humans (Wikel 1999). Because ticks are blood-feeding arthropods, they can cause severe anaemia in their hosts; infestation with about 200 ticks for more than 6 weeks may be fatal (Frisch *et al.* 2000). The long duration of blood-feeding by the hard ticks (e.g. *Riphicephalus* spp.), unique among ectoparasites, induces haemostatic, inflammatory and immune responses in the host (Hajnická *et al.* 2005). To be successful in their feeding task, ticks manipulate their host's immune system via several substances in their saliva, such as antihaemostatic, anti-inflammatory and immunomodulatory agents (Brossard & Wikel 2004). It has been shown that tick salivary gland extracts can promote virus growth *in vitro* (Hajnická *et al.* 1998).

Ticks are usually controlled with accaricides, which are not totally effective (Graf *et al.* 2004), and there are increasing concerns about environmental safety and human health with the use of accaricides. Biological control could be an alternative approach because ticks have numerous natural enemies, but only a few species have been evaluated as potential tick biocontrol agents (Samish *et al.* 2004).

Although cattle are able to develop an immune response after successive exposures to ticks (Dossa *et al.* 1996), vaccine development has not been totally successful because of the great ability of parasites to modulate the host immune system (Brossard & Wikel 2004), probably because the parasites attack their hosts through interaction with the host cytokines (Wikel & Alarcon-Chaidez 2001).

Based on this information, breeding for resistance is still considered one of the best alternatives to control tick infestation in cattle. Resistance to ticks is a trait that has a moderate heritability. Davis (1993) found an average heritability of 0.34 ± 0.06, similar to the heritability of milk production. Henshall (2004) found values of 0.41 ± 0.08 for mean (log) tick count in *B. taurus* breeds. Although there is genetic variability among individuals and breeds (Morris 2006), the identification of superior genotypes is a difficult task. However, Acosta-Rodriguez *et al.* (2005) and Martinez *et al.* (2006) found associations between some BoLA class II microsatellite alleles and susceptibility to *R. microplus* ticks in cattle.

Because *B. indicus* cattle are generally more resistant to parasite challenge than are *B. taurus* (Wambura *et al.* 1998), we exploited the genetic variability between these subspecies to map QTL for resistance to the *R. microplus* tick in an F_2 (Holstein × Gir) experimental population. In this study, we report results from QTL analysis of bovine chromosomes 5, 7 and 14. We selected these chromosomes for our initial analysis because genes related to host responses to parasite are located on these chromosomes: *IFNG* on BTA5, *IL3*, *IL4* and *IL5* on BTA7 and *IL7* on BTA14.

Materials and methods

Resource population

Using multiple ovulation followed by embryo transfer, a Holstein × Gir population of 382 F_2 animals was developed on an experimental farm located in Valença (22°15′S, 43°41′W), state of Rio de Janeiro, Brazil. The experimental area is hilly, with altitudes between 200 and 400 m above sea level and a climate corresponding to Cwa of Koppen's classification (Koppen & Geiger 1936). From 1999 to 2005, five F_1 sires and 68 F_1 dams were obtained from crosses of four Holstein sires × 28 Gir dams, and intercrossed to produce the F_2 generation.

Phenotypic measurements

The F_2 calves were reared as described in Martinez *et al.* (2006). Tick exposure was continuous under natural conditions with no tick control until 10–14 months of age, when animals were evaluated for tick resistance by experimental challenge using contemporary groups of 20–30 animals. Absolute tick count was determined by counting the female ticks that completed the biological cycle after artificial infestations with about 10 000 *R. microplus* larvae per animal. These infestations were carried out by placing tick larvae in the dorsal–lumbar region of the animals. Animals were kept tied up for 30 min to prevent grooming and to allow the larvae to spread to all regions of the body. Counts were made on the morning of the 21st day after infestation, before the engorged ticks detached from the animals. All engorged female ticks (4.5–8.0 mm in length) were counted on one side of the animal, and the count was then multiplied by 2.0. Ticks were counted in the rainy season (October–March; $N = 302$) and the dry season (April–September; $N = 338$).

Marker selection and genotyping

Microsatellite markers were selected from the USDA, ARS, Meat Animal Research Center (USMARC) database at http://www.marc.usda.gov, based on chromosome position, allele number, polymorphic information content (PIC), allele range, annealing temperature and, most importantly, heterozygosity level in the F_1 bulls. PCR conditions were described in Gasparin *et al.* (2005), and amplified products were analysed using an ABI Prism 3100

Avant sequencer (Applied Biosystems). Results were analysed using GENESCAN 3.7 and GENOTYPER 3.7.1 software.

Statistical analysis

Linkage maps were assembled using the BUILD and ALL functions of the CRIMAP program (Green *et al.* 1990), and the parental contribution of each locus was determined using the CHROMPIC function, which, starting from the genotypes of a three-generation pedigree (parental, F_1 and F_2), allowed determination of the linkage phases of the markers and the haplotypes in the F_2 generation. The maps were derived from the observed recombination fraction for each marker interval using Kosambi's mapping function to transform recombination into distance.

In the QTL analysis, effects of sex, year in which the animal was evaluated, coat colour (totally white, more than 75% white, 50–75% white and <50% white) and hair type (short straight, short curly, long straight and long curly) were included as fixed effects. The age of the animal at evaluation (days) was included as a covariable. Tick counts were normalized using a ln(absolute tick count + 1) transformation. The transformed data, hereafter called LogTick, was submitted to a normality test using BIOESTAT 2.0 (Ayres *et al.* 2000). Significance of the fixed effects and covariables for each trait was evaluated by analysis of variance using the general linear model (GLM) procedures of SAS software (SAS Institute 2002).

The maps were then used for QTL analysis by the regression interval mapping method for F_2 families (Haley *et al.* 1994) using the QTL EXPRESS program at http://QTL.cap.ed.ac.uk (Seaton *et al.* 2002). The *F*-statistic was calculated to test the hypothesis of QTL segregation at 1-cM intervals using a model that included all the fixed effects that were significant and the additive and dominance effects of the QTL. Permutation was used to determine the threshold significance (Churchill & Doerge 1994), and the bootstrap technique was used to determine the confidence interval (CI) of the QTL (Visscher *et al.* 1996). In this study, 10 000 permutations were used to obtain stable estimates for the value of $\alpha = 0.01$ (Churchill & Doerge 1994).

Results and discussion

Phenotypic measurements

Tick counts in the rainy season averaged 40 ± 72.4, with eight animals showing more than 200 ticks. The maximum observed value was 792 ticks in one animal. Twenty-five animals were free of ticks in the rainy season. The dry season had an average of 33 ± 43.3 ticks per animal. The maximum observed value was 412 ticks; four animals had more

Table 1 Influence of year of evaluation, gender, coat colour and hair type over tick count in each season.

	Rainy season			Dry season		
	N	Average[1]	Standard error	*N*	Average[1]	Standard error
Year						
2001	22	29.72 (2.8)	31.05 (1.3)	46	18.35 (2.47)	19.73 (1.08)
2002	34	63.82 (3.67)	61.4 (1.15)	71	34.17 (3.01)	39.46 (1.11)
2003	116	36.07 (3.16)	41.87 (1.02)	26	29 (2.98)	25.05 (1.1)
2004	1	–	–	90	36.42 (3.13)	43.49 (0.98)
2005	65	23.69 (2)	43.28 (1.6)	67	38.8 (2.85)	64.21 (1.35)
2006	64	80.47 (3.83)	130.97 (0.97)	38	31.58 (3.12)	32.41 (0.83)
Sex						
Male	149	45.98 (3.19)	65.53 (1.28)	166	36.77 (3.1)	49.02 (1.09)
Female	152	45.12 (3)	83.26 (1.4)	168	29.34 (2.83)	37.17 (1.12)
Coat colour[2]						
1	12	20.83 (2.54)	15.41 (1.4)	46	17.48 (2.44)	18.24 (1.03)
2	65	33.44 (3.06)	34.82 (1.05)	87	34.11 (2.98)	44.38 (1.1)
3	114	49.31 (3.25)	62.66 (1.39)	95	33.32 (2.96)	52.35 (1.07)
4	111	51.11 (3)	101.7 (1.44)	110	37.85 (3.13)	40.41 (1.13)
Hair type[3]						
A	204	32.67 (2.86)	34.99 (1.34)	141	22.35 (2.6)	28.38 (1.1)
B	49	83.02 (3.71)	142.31 (1.13)	89	36.67 (3.14)	42.27 (1.06)
C	45	55.6 (3.37)	69.96 (1.31)	68	37.23 (3.13)	39.73 (1.06)
D	4	120 (3.9)	198.73 (1.51)	40	53.85 (3.43)	75.48 (1.03)

[1]Transformed values [$\log_n$(number of ticks + 1)] are in parentheses.

[2]1 = totally white; 2 = more than 75% white; 3 = 50–75% white; 4 = <50% white.

[3]A = short straight; B = short curly; C = long straight; D = long curly.

than 200 ticks and 10 animals were completely free of ticks. Environmental effects differed between the seasons. Year of evaluation and hair type were significant for tick counts during the rainy season ($P < 0.01$). For the dry season, tick count was affected by coat colour and hair type ($P < 0.01$).

Animals with long and curly hair had two times more ticks than animals with short and straight hair (Table 1). This tendency has also been shown by Fraga *et al.* (2003) and Martinez *et al.* (2006). Long hair makes self-cleaning against ticks more difficult for cattle (Fraga *et al.* 2003). Also, animals with fur coverage appropriate for tropical weather (thinner coats) suffer less thermal stress and, consequently, have a better immunological response (Hansen 2004). Thermal stress may impair tick resistance, explaining why darker-coat-colour animals showed more ticks (Table 1), similar to the results of Fraga *et al.* (2003) as well.

Although Stear *et al.* (1990) and Martinez *et al.* (2006) observed significant effects of animal gender on resistance to ticks, which they attributed to hormone differences, the sex effect was not significant in our data set, and it was not included in the final QTL analysis model.

Data normalization after logarithmic transformation was significant ($P < 0.05$) using the D'Agostino normality test (Baringhaus & Henze 1990).

Genotyping and linkage maps

Most markers were considered highly polymorphic, with heterozygosities (H) ≥ 0.7 (Ott 1992) and an average PIC of 0.645 (Table 2). In a study of three *B. indicus* dairy breeds with 20 microsatellites, Mukesh *et al.* (2004) found an average PIC value of 0.610. The similarity between these PIC averages may reflect the restricted number of founders in the F_2 population.

Linkage maps of the three chromosomes constructed in this study generally agreed with the MARC reference maps. However, distances between the markers were larger than predicted, and an inversion between *BMS1617* and *BMS490* on BTA5 was found (Table 2). This was probably due to the lack of coinformative meioses for some markers, leading to less-precise mapping of the markers (Liu 1998).

Table 2 Summary information for microsatellite markers used in the study.

				Map position (cM)		Informative meioses	
Chromosome	Number of alleles	Heterozygosity (H)	PIC	MARC	This study	MARC	This study
BTA5							
BM6026	6	0.842	0.715	6.05	0.0	445	678
BP1	3	0.535	0.425	17.29	19.5	2810	317
BM321	7	0.850	0.751	38.24	54.2	1230	657
BMS1617	7	0.679	0.625	56.30	93.9	321	499
BMS490	6	0.882	0.745	66.20	77.4	3454	440
BMS1248	6	0.679	0.592	90.85	132.2	259	502
ILSTS034	10	0.808	0.777	103.44	148.0	322	622
ETH152	6	0.591	0.602	121.75	169.5	3016	445
BTA7							
INRA192	7	0.685	0.673	0.0	0.0	2522	606
BM9065	4	0.733	0.659	29.61	32.9	1484	479
ILSTS006	4	0.711	0.597	32.04	48.7	4727	457
BM7160	4	0.711	0.613	32.04	49.5	1673	549
BM2607	3	0.664	0.577	62.24	93.1	156	550
BOBT24	5	0.777	0.715	82.48	122.6	261	601
IL4	5	0.777	0.712	101.11	155.6	3620	601
BM6117	3	0.335	0.329	116.63	199.0	558	207
BTA14							
CSSM066	7	0.845	0.824	5.0	0.0	241	562
ILSTS011	3	0.665	0.590	10.6	16.9	265	535
BMC1207	4	0.434	0.385	36.2	47.8	839	234
BMS740	6	0.826	0.800	44.2	67.7	422	598
BMS1899	6	0.781	0.747	72.0	91.6	363	561
BL1036	8	0.784	0.757	78.7	116.8	249	536
BMS2055	4	0.674	0.617	84.1	144.3	422	415
Mean	5.39	0.707	0.645			1290	507

PIC, polymorphism information content; MARC, Meat Animal Research Center (http://www.marc.usda.gov/).

QTL mapping for tick resistance

In the first QTL model tested, only the additive QTL effect was considered. For LogTick during the rainy season, a chromosome-wide significant QTL ($P < 0.01$) for tick load was detected on BTA5 ($F = 13.06$), one suggestive QTL ($P < 0.10$) was found on BTA7 ($F = 6.37$) and no QTL were found on BTA14. When LogTick data from the dry season were analysed, no significant QTL were detected on BTA5 ($F = 4.84$), one suggestive QTL ($P < 0.05$) was found on BTA7 ($F = 9.0$) and a chromosome-wide significant QTL ($P < 0.01$) was mapped to BTA14 ($F = 13.01$).

In the second QTL analysis, both additive and dominance effects were considered, and the significance of dominance deviation for each QTL was assessed using an *F*-test of the two models. Dominance deviation was significant for LogTick QTL mapped on BTA5 for the rainy season and LogTick QTL at the dry season on BTA14 ($P < 0.01$). The QTL for LogTick at the rainy and dry seasons on BTA7 also had a significant dominance deviation ($P < 0.05$). These results suggest that these QTL are influenced not only by the QTL allele effect itself, but also by the combination of alleles.

Including dominance effects in the model of the rainy data set reduced the significance of the 1% chromosome-wide significant QTL on BTA5 to 5% ($F = 6.63$; Fig. 1). The 10% significance level of BTA7 QTL disappeared (Fig. 2), and no QTL were found on BTA14 (Fig. 3). Including dominant deviation in the analysis of the dry season resulted in no QTL on BTA5, the 5% significance threshold QTL of BTA7 decreased to 10% ($F = 4.52$) and one suggestive QTL ($P \leq 0.05$) was found on BTA14 ($F = 6.53$). The positions of the QTL on the chromosomes, significance levels, *F*-statistics values, LOD scores and CI are summarized in Table 3.

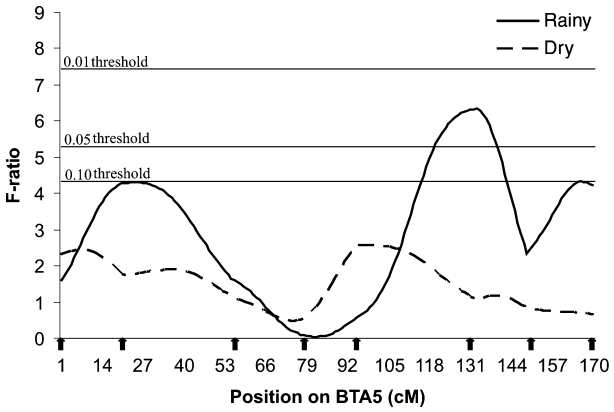

Figure 1 *F*-statistic profile for LogTick evaluated during rainy and dry seasons on BTA5. The horizontal lines represent the significant thresholds. Arrows on the *x*-axis indicate the relative position of markers *BM6026, BP1, BM321, BMS1617, BMS490, BMS1248, ILSTS034* and *ETH152*.

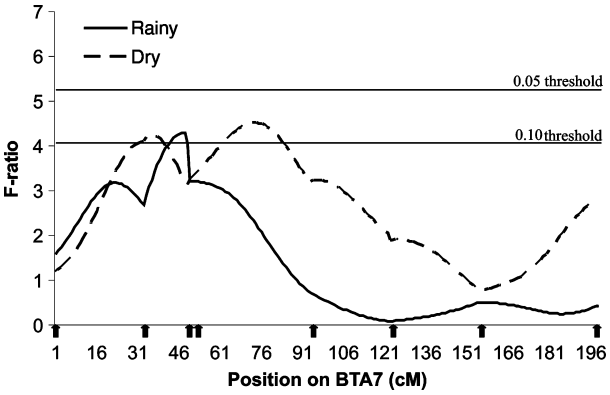

Figure 2 *F*-statistic profile for LogTick evaluated during rainy and dry seasons on BTA7. The horizontal lines represent the significant thresholds. Arrows on the *x*-axis indicate the relative position of markers *BM7160, BMS2607, BOBT24, IL4, BM6117, INRA192, BMS9065* and *ILSTS006*.

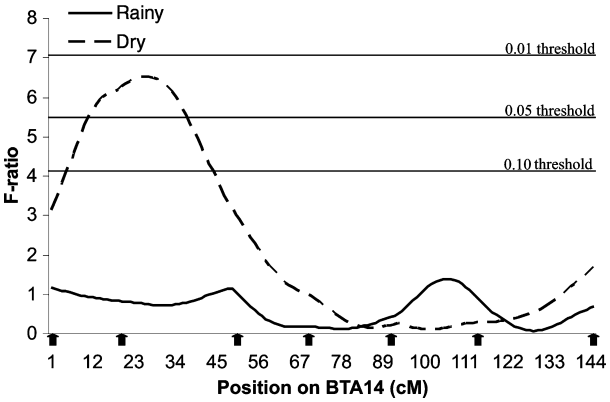

Figure 3 *F*-statistic profile for LogTick evaluated during rainy and dry seasons on BTA14. The horizontal lines represent the significant thresholds. Arrows on the *x*-axis indicate the relative position of markers *CSSM066, ILSTS011, BMC1207, BMS740, BMS1899, BL1036* and *BMS2055*.

Table 3 QTL for tick resistance identified on BTA5, BTA7 and BTA14 during the rainy and dry seasons.

	LOD	Significance (%)	Position (cM)	*F*-ratio	CI (cM)
Rainy					
BTA5	2.815	5	132	6.63	10–169
BTA7	1.835	NS	47	4.29	3–198
BTA14	0.603	NS	105	1.4	0–144
Dry					
BTA5	1.107	NS	96	2.57	0–169
BTA7	1.935	10	73	4.52	9.5–198
BTA14	2.770	5	25	6.53	0–144

NS, not significant ($P > 0.10$); CI, confidence interval.

We also examined the amount of phenotypic variance that was explained by the additive effect of the QTL. On BTA5, the additive effect found in the rainy season corresponded to only 1.7% of the total phenotypic variance, while on BTA7 and BTA14 the additive effect corresponded to 1.9 and 3.2% respectively.

Ashton *et al.* (1968) found an association between serum amylase phenotype, which is located on BTA3, and tick infestation in cattle. A similar result was found by Panepucci *et al.* (1989) in a study with Canchim cattle (3/8 Zebu, 5/8 Charolais). Acosta-Rodriguez *et al.* (2005) analysed microsatellite markers in the bovine BoLA class II complex and found associations between two of these microsatellites and tick infestation. Martinez *et al.* (2006) found an association between BoLA alleles and tick number ($P < 0.05$) in the same F_2 population used in this research.

Environmental and physiological factors can alter tick counts in cattle. Ambient temperature and humidity are important factors that influence the number of ticks that develop to the adult phase on an animal (Powell & Reid 1982). This could explain why tick counts were different between the seasons and why the QTL effect and localization changed when analysing these seasons separately. Coat colour is another important factor for resistance against parasites, because darker animals may be less immunologically competent under the heat of the tropical countries because of thermal stress (Hansen 2004).

Because the additive effect is half the difference between parental QTL genotypes, positive values in our study indicate that the allele that increases tick loads originates from Holstein. The additive effect of the BTA5 QTL for the rainy season had a negative value, indicating that the resistant QTL alleles originated from Gir. On the other hand, QTL for the dry season on BTA7 and BTA14 had positive additive values, with resistant alleles from the Holstein breed. In Brazil, the rainy season coincides with the warmer summer and spring and the dry periods occur mainly during the colder seasons of the year. The different parental origins of the QTL across the seasons could be explained by the adaptability of *B. indicus* to the tropical weather, *B. taurus* being more effective against parasite infestation in lower temperatures.

The QTL in question may depend on environmental factors. If these effects are ignored in the mapping model, the QTL detection power may be reduced and the resulting estimates may be biased. Genotype × environment interaction or differential genotypic expression across environments can reduce the association between phenotypic and genotypic values, causing selected animals from one environment to perform poorly in another. Moreover, the sampling problem associated with seasonal variation suggests that the analysis across multiple years is necessary.

The CIs in this study are extremely large (in cM) and span nearly the entire length of each chromosome. Once the interval is reduced by adding more markers to the analysis, other experimental populations should be evaluated to validate the effects of these regions on tick infestation.

Acknowledgements

We thank the Embrapa Dairy Cattle Research Centre staff for all the phenotypic data. This work was financially supported by Prodetab. We thank CNPq for providing scholarships to GG and MM and fellowships to LLC, MLM and LCAR.

References

Acosta-Rodriguez R., Alonso-Morales R., Balladares S., Flores-Aguilar H., Garcia-Vazquez Z. & Gorodezky C. (2005) Analysis of BoLA class II microsatellites in cattle infested with *Boophilus microplus* ticks: class II is probably associated with susceptibility. *Veterinary Parasitology* **127**, 313–21.

ANUALPEC (2005) *Anuário da Pecuária Brasileira*. Instituto FNP, São Paulo.

Ashton G.C., Seifert G.W. & Francis J. (1968) An association between serum amylase phenotype and tick infestation in cattle. *Australian Journal of Biological Sciences* **21**, 303–8.

Ayres M., Ayres M. Jr, Ayres D.L. & Santos A.S. (2000) *BioEstat 2.0: Aplicações estatísticas nas áreas de Ciências Biológicas e Médicas*. Sociedade Civil Mamirauá, CNPq, Brasília.

Baringhaus L. & Henze N. (1990) A test for uniformity with unknown limits based on D'Agostino's. *Statistics & Probability Letters* **9**, 299–304.

Brossard M. & Wikel S.M. (2004) Tick immunobiology. *Parasitology* **129**, S161–76.

de Castro J.J. (1997) Sustainable tick and tick-borne disease control in livestock improvement in developing countries. *Veterinary Parasitology* **71**, 77–97.

Churchill G.A. & Doerge R.W. (1994) Empirical threshold values for quantitative trait mapping. *Genetics* **138**, 963–71.

Davis G.P. (1993) Genetic parameters for tropical beef cattle in Northern Australia: a review. *Australian Journal of Agriculture Research* **44**, 179–98.

Dossa S.D., Kaaya G.P., Essuman S., Odulaja A. & Assoku R.G.K. (1996) Acquisition of resistance tick *Amblyomma variegatum* in Boran cattle, *Bos indicus* and the effects of *Trypanosoma congolense* and *Babesia bigemina* on host resistance. *Veterinary Parasitology* **62**, 317–30.

Fraga A.B., Alencar M.M., Figueiredo L.A., Razook A.G. & Cyrillo J.N.S.G. (2003) Análise de fatores genéticos e ambientais que afetam a infestação de fêmeas bovinas da raça Caracu por carrapatos (*Boophilus microplus*). *Revista Brasileira de Zootecnia* **32**, 1578–86.

Frisch J.E., O'Neill C.J. & Kelly M.J. (2000) Using genetics to control cattle parasites – the Rockhampton experience. *International Journal of Parasitology* **30**, 253–64.

Gasparin G., Miyata M., Coutinho L.L., Martinez M.L., Machado M.A., Silva M.V.G.B., Campos A.L. & Regitano L.C.A. (2005) Quantitative trait locus affecting birth weight on bovine chromosome 5 in a F_2 Gyr × Holstein population. *Genetics and Molecular Biology* **28**, 670–6.

Graf J.F., Gogolewski R., Leach-Bing N., Sabatini G.A., Molento M.B., Bordin E.L. & Arantes G.J. (2004) Tick control: an industry point of view. *Parasitology* **129**, S427–42.

Green P., Falls K. & Crooks S. (1990) *Documentation for CRIMAP, Version 2.4*. Washington University School of Medicine, St Louis, MO, USA.

Hajnická V., Fuchsberger N., Slovak M., Kocakova P., Labuda P. & Nuttall P.A. (1998) Tick salivary gland extracts promote virus growth in vitro. *Parasitology* **116,** 533–8.

Hajnická V., Vancova I., Kocakova P., Slovak M., Gasparik J., Slavikova M., Hails R.S., Labuda M. & Nuttall P.A. (2005) Manipulation of host cytokine network by ticks: a potential gateway for pathogen transmission. *Parasitology* **130,** 333–42.

Haley C.S., Knott S.A. & Elsen J.M. (1994) Mapping quantitative trait loci in crosses between outbred lines using least squares. *Genetics* **136,** 1195–207.

Hansen P.J. (2004) Physiological and cellular adaptations of zebu cattle to thermal stress. *Animal Reproduction Science* **82,** 349–60.

Henshall J.M. (2004) A genetic analysis of parasite resistance traits in a tropically adapted line of *Bos taurus*. *Australian Journal of Agricultural Research* **55,** 1109–16.

Horak I.G., Camicao J.L. & Keirans J.E. (2002) The Argasidae, Ixodidae and Nuttalliellidae (Acari: Ixodida): a world list of valid tick names. *Experimental and Applied Acarology* **28,** 27–54.

Jongejan F. & Uilenberg G. (2004) The global importance of ticks. *Parasitology* **129,** 3–14.

Koppen W. & Geiger R. (1936) *Handbuch der Klimatologie*, Vol. 1. Verlag, Börntraeger, Berlin, Germany.

Liu B.H. (1998) *Statistical Genomics: Linkage Mapping and QTL Analysis*. CRC Press, Boca Raton, FL.

Martinez M.L., Machado M.A., Nascimento C.S. *et al.* (2006) Association of BoLA-DRB3.2 alleles with tick (*Boophilus microplus*) resistance in cattle. *Genetics and Molecular Research* **5,** 513–24.

Morris C.A. (2006) A review of genetic resistance to disease in *Bos taurus* cattle. *The Veterinary Journal* (doi:10.1016/j.tvjl.2006.09.006, in press).

Mukesh M., Sodhi M., Bhatia S. & Mishra B.P. (2004) Genetic diversity on Indian native cattle breeds as analysed with 20 microsatellite markers. *Journal of Animal Breeding and Genetics* **121,** 416–24.

Ott J. (1992) Strategies for characterizing highly polymorphic markers in human gene mapping. *American Journal of Human Genetics* **51,** 283–90.

Panepucci L.L.L., Alencar M.M. & Oliveira G.P. (1989) Estudo preliminar da relação entre marcadores genético-bioquímicos e resistência ao carrapato em gado de corte. *Revista da Sociedade Brasileira de Zootecnia* **18,** 172–8.

Powell R.T. & Reid T.J. (1982) Project tick control. *Queensland Agricultural Journal* **108,** 279–300.

Samish M., Ginsberg H. & Glazer I. (2004) Biological control of ticks. *Parasitology* **129,** S389–403.

SAS Institute (2002) *SAS/STAT User's Guide*, Version 8.0, 5th edn. SAS Institute, Inc., Cary, NC, USA.

Seaton G., Haley C.S., Knott S.A., Kearsey M. & Visscher P.M. (2002) QTL Express: mapping quantitative trait loci in simple and complex pedigrees. *Bioinformatics* **18,** 339–40.

Stear M.J., Hetzel D.J., Brown S.C., Gershwin L.J., Mackinnon M.J. & Nicholas F.W. (1990) The relationships among ecto- and endoparasite levels, class I antigens of the bovine major histocompatibility system, immunoglobulin E levels and weight gain. *Veterinary Parasitology* **34,** 303–21.

Visscher P.M., Thompson R. & Haley C.S. (1996) Confidence intervals in QTL mapping by bootstrapping. *Genetics* **143,** 1013–20.

Wambura P.N., Gwakisa P.S., Silayo R.S. & Rugaimukamu E.A. (1998) Breed-associated resistance to tick infestation in *Bos indicus* and their crosses with *Bos taurus*. *Veterinary Parasitology* **77,** 63–70.

Wikel S.K. (1999) Tick modulation of host immunity: an important factor in pathogen transmission. *International Journal of Parasitology* **29,** 851–9.

Wikel S.K. & Alarcon-Chaidez F.J. (2001) Progress toward molecular characterization of ectoparasite modulation of host immunity. *Veterinary Parasitology* **101,** 275–87.