Genetic Adaptation of Livestock to Heat Stress Challenges

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Contents

Abstract

Genetic/genomic selection between and within species and breeds can aid in maintaining production levels in farm animal species under climatic stress. This chapter looks at how genes and animals can be identified and used for this purpose. We also look at over 19,600 genes reported from studies on adaptation cited in the scientific literature for cattle, sheep, goats and horses. Functional analysis revealed pathways involved in developmental and growth processes, regulation (positive and negative) of biological process, regulation of response to stimulus and stress, immune system regulation, function and development, leukocyte activation, oxidoreductase activity, metabolism and behaviour. Future works will look at how we can select for increased tolerance to heat stress and its related traits while maintaining productivity. Solutions may include landscape genomics, genome editing and multi-omics studies. Overall, there is a need to integrate different stakeholders with the development of statistical methodologies

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V. Sejian et al. (eds.), *Climate Change and Livestock Production: Recent Advances and Future Perspectives*, https://doi.org/10.1007/978-981-16-9836-1_21

(including artificial intelligence and machine learning) and a regulatory framework to ensure animal welfare and consumer safety.

Keywords

Gene ontology · GWAS · Landscape · Selection · Sequencing

Abbreviations

B4GALT6	Beta-1,4-galactosyltransferase 6
Cas9	CRISPR associated protein 9
CRISPR	Clustered regularly interspaced short palindromic repeats
DNA	Deoxyribonucleic acid
DSC	Desmocollins
DSG	Desmogleins
EGFR	Epidermal growth factor receptor
FDR	False discovery rate
Fig	Figure
F_{ST}	Fixation index
GEBV	Genomic breeding value
GO	Gene ontology
GWAS	Genowe-wide association study
HS	Heat stress
KEGG	Kyoto Encyclopedia of genes and genomes
LISA	Local indicators of spatial association
MAPK	Mitogen-activated protein kinase
mTOR	mammalian target of rapamycin
PI3k/AKT	phosphatidylinositol 3-kinase/protein kinase B
QTL	Quantitative trait loci
RNA	Ribonucleic acid
SNPs	Single nucleotide polymorphisms
TTR	Transthyretin

21.1 Introduction

There is an increased risk of extreme heat stress (HS) in domestic animals due to climate change, especially in the tropics. Animals undergo stress (internal or external stimuli that disrupt homeostasis) in all types of production systems. Still, how the organism responds to this determines if it remains healthy and productive. This response can be at several levels, including physiological, cellular, molecular or behavioural. Nevertheless, to understand how stress affects an organism, it is necessary to comprehend how response can be affected by several complex and overlapping factors.

Heat stress does not typically occur in isolation. It can be accompanied by a lack of water and feed (Sejian et al. 2013), the need to walk long distances, increased or decreased humidity, and disease/vector prevalence. Input constraints such as forage availability and quality, feed digestion and absorption occur in sequence. Therefore, effects may be directly (thermoregulation, effects on endocrine, metabolism, production, and reproduction systems) or indirectly (environmental effects on food and water supply, pest and pathogen prevalence, and immune system resistance immunological pressures) affect the animal. Under HS, as described above, animals reduce food intake and, thereby, digestive capacity. Consequently, fewer nutrients are absorbed. Reproduction is compromised, followed by production and growth, with energy conserved for vital maintenance functions.

In animal production, selective breeding has improved productivity using modern animal breeding techniques and reproductive technologies. The response is clear with marked increases in efficiency and production levels but with a reduction in fitness, resistance to disease and tolerance to increasing stresses in the environment such as higher temperatures or lower water availability. Pryce and Haile-Mariam (2020) show that heat tolerance has worsened in dairy cattle due to a lack of selection for these traits. Climatic variables determine the choice of livestock species after soils, geography, household characteristics, and country fixed effects have been considered. An individual's susceptibility to heat stress depends on both intrinsic (mainly of genetic origin) and extrinsic (primarily environmental) factors (Gaughan et al. 2002). Also, traits that were not previously deemed relevant, such as methane production, have become important for the livestock industry (Hayes et al. 2013), as well as the need to meet consumer demands for healthier animal products.

Genetic selection is incorporated into breeding programs based on linkage-disequilibrium between polymorphisms in relevant characteristics and markers (usually SNPs – Single Nucleotide Polymorphisms). Genomic breeding values (GEBV) are calculated as the sum of each genetic marker's effects or haplotypes. As these are spread across the entire genome, potentially all quantitative trait loci (QTL) that contribute to variation in the trait of interest are captured. For this, a reference population is used whereby animals are both phenotyped and genotyped for the characteristics of interest. Hayes et al. (2009) showed that linkage disequilibrium varied between breeds. When the prediction equations are to be used across multiple breeds, the definition of the optimal composition of reference populations is necessary.

Genomic selection can aid in selection schemes, as traits related to the response to HS may be challenging to measure or have low heritability. This depends on sufficient phenotyping on reference populations to determine linkages between genotypes and phenotypes. Another option is the identification of genes of large effects, such as the slick hair gene in locally adapted cattle breeds (Olson et al. 2003). Dikmen et al. (2014) showed that introgression of this gene into Holstein cattle improved thermoregulatory ability, with positive effects on milk yield even under conditions whereby housing was modified to reduce HS. More recently, genome editing (especially using CRISPR-Cas9 technology) has been used to introduce beneficial alleles (e.g., heat tolerance, disease resistance) and haplotypes from native

locally adapted to commercial populations to improve their productivity under climate change (Singh and Ali 2021).

While mitigation of the effects of climate change on livestock through modification of management conditions may represent a short-term solution to heat stress challenges, genetic changes are more long term. Studies with locally adapted breeds can help identify genes and metabolic pathways of interest in adaptation studies, as they have had sufficient time to become genetically adapted to the environment. These adaptations arise through equilibrium between evolutionary (crossbreeding, artificial selection, genetic drift) and local environment sources.

Collier et al. (2019) identified three responses of the animal to HS: acclimation (phenotypic response to a specific stressor), acclimatisation (coordinated response to more than one stressor) and adaptation (genetic modification as adverse environment persists over a few generations). Therefore, coping involves (McManus et al. 2020) the ability to limit the heat load (resistance) and the ability to limit the harm caused by a specific stressor (tolerance).

The animal's response tries to minimise the effects of HS on cellular functions. It does this through a coordinated gene network involving various cells, tissues and systems. These genes are involved in (McManus et al. 2020) energy production/metabolism, classical heat shock protein genes/chaperones, protein degradation/turnover/DNA repair, genes reducing the impact of oxidative stress/cellular repair, and transcriptional regulation. Gene expression is a crucial component of the cellular response to thermal stress.

21.2 Methods for Identifying Specific Genes or Alleles for Adaptation

Adaptation traits are complex and controlled by many genes control. These genes are commonly grouped based on function or common expression profiles. This grouping attempts to summarise a complex response network into fewer categories. These response categories can then be interpreted due to their perceived roles in re-establishing cellular homeostasis.

Molecular genetic approaches (such as microarray analyses, whole transcriptome analysis, genome-wide association studies and next-generation sequencing) have been used to identify adaptation-related genes. These, in turn, have been used to group the genes associated with thermo-tolerance into different categories. Genome-wide association studies (GWAS) use genetic variations in the genotype and phenotype by scanning the genomes of many different animals and looking for those statistically associated with a specific trait or disease. This method aims to identify common single nucleotide polymorphisms (SNPs) in the animal genome and determine how these polymorphisms are distributed across different populations. Other methods analyse the patterns of genomic diversity within and between populations, as well as the level of admixture in specific genomic regions, to identify adaptive selection signatures. Landscape Genomics use environmental variables as proxies

for phenotypes. These approaches require genomic tools to look at individual loci via whole genomic sequence analyses.

HS has been shown to stimulate signal transduction pathways. This stimulus alters gene expression of immune cell mediators, thereby activating the heat shock response, promoting cytokine activity. In sequence, this impairs the cellular immune response by increasing cortisol concentrations, which binds to a specific Transcription Factor that control gene function. Physiological responses are linked to these molecular pathways and processes. Adaptive studies quantify responses through mechanisms such as gene/protein expression, enzyme activity and genome-wide analyses. For these studies to be feasible, there must be sufficient genetic variation within a population.

21.3 Genes Linked to Heat Stress

Vertebrate genomes have millions of single nucleotide polymorphisms (SNPs). Genetic variation can be due to deletions, duplications, copy-number variants, insertions, inversions and translocations, mobile genetic elements, splicing junction heterogeneity, regulatory elements and different sorts of ploidies. On the other hand, crossing over, independent assortment and sexual reproduction are principal mechanisms that maintain genetic diversity within populations. Genotype-environment interactions cause phenotypic variation, which provides the substrate for adaptive mechanisms.

We used three online tools (http://bioinformatics.sdstate.edu/go/; https://tools.dice-database.org/GOnet/ and https://david.ncifcrf.gov/tools.jsp) to analyse over 19,906 genes (Fig. 21.1) found in published papers on the adaptation of cattle, sheep, goats and horses worldwide.

As can be seen, most genes were cited in articles on cattle, and all species showed significant overlaps. The genes found in at least three of the four species were then used in further analyses (1703 genes used for enrichment, KEGG pathways, gene ontology analyses).

Functional analysis revealed pathways involved in developmental and growth processes, regulation (positive and negative) of biological process, response to stimulus and stress, immune system regulation, process and development, leukocyte activation, oxidoreductase activity, metabolism and behaviour (Fig. 21.2). As can be seen, the response to HS involves several response levels. As the papers from which these genes were identified are globally distributed and responses may be region or breed-specific, further studies are needed to improve our understanding of these processes.

Enrichment analysis (Fig. 21.3) shows 19 enriched regions (Bos taurus used as the reference genome for the four species) on chromosomes 3, 7, 8, 13, 18, 19 and 24. For example, the enriched region on chromosome 24 shows genes linked to epidermal, epithelial and mucosal structures (DSC1, DSC2, DSC3, DSG1, DSG2, DSG3, DSG4), thyroxine transport (TTR), glycolipid biosynthesis (B4GALT6), and protein modification (GALNT1), among others.

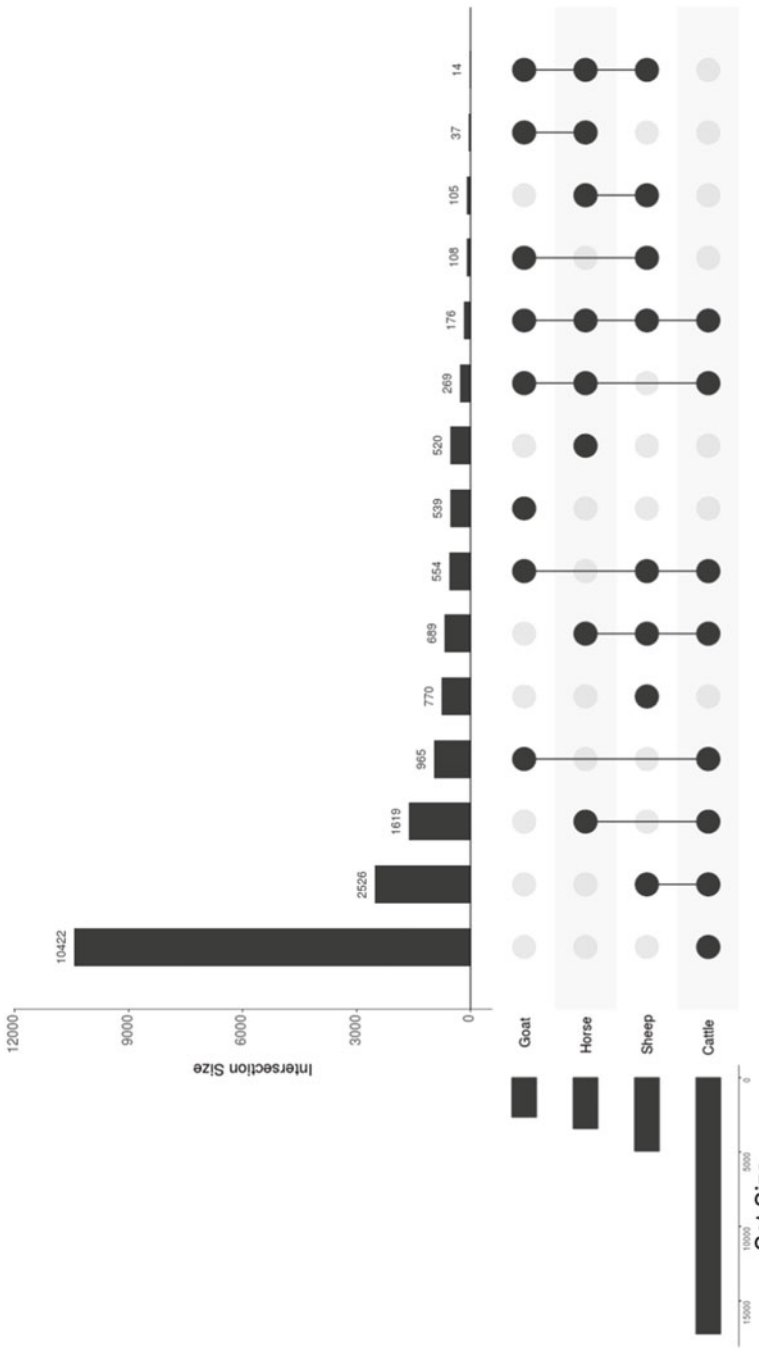

Fig. 21.1 Upset Plot (https://gehlenborglab.shinyapps.io/upsetr/) of the number of genes linked to climatic adaptation cited in scientific papers per species (based on 19,609 genes and 127 scientific papers)

Fig. 21.2 Functional analysis network from 1703 genes related to heat response in cattle, sheep, goats and horses (https://tools.dice-database.org/GOnet/)

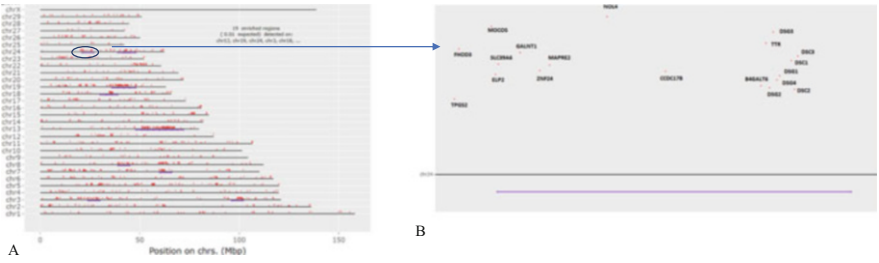

Fig. 21.3 Chromosomal localization of 1703 genes found in published papers on the adaptation of cattle, sheep, goats and horses worldwide (**a**) and a closeup (**b**) of one of the enriched regions based on cattle genomic organization (individual genes in red, enriched regions underlined in purple - these regions were defined by chi-square tests using all 19,906 genes found in the literature. http://bioinformatics.sdstate.edu/go/)

Signalling networks, such as MAPK (immune response, gene expression, cell proliferation, differentiation, apoptosis, and cell motility), mTOR (growth, energy metabolism, ageing), ErbB (cell growth, development and survival, lipid metabolism), and PI3k/AKT (metabolism, proliferation, cell survival, growth and angiogenesis) are also involved in this response (Fig. 21.4 and Table 21.1).

In the reactome (https://reactome.org/) analysis, the importance of the immune response can be seen due to the prevalence of interleukin and MAPK signalling. Here too, the response to heat stress is highlighted. AKT (serine/threonine kinase) is an upstream positive regulator of the mammalian target of rapamycin (mTOR) which controls cell growth. The KEGG analyses (https://www.genome.jp/kegg/pathway.html) highlights some of the same reactions, but also includes melanogenesis and epidermal growth factor receptor (EGFR) tyrosine kinase inhibitor resistance possibly involved in primary defence of the organisms to ultraviolet light. Rap1 regulates the cell's response to external stimuli, and is also involved in cell survival and proliferation. In the Gene Ontology (http://geneontology.org/) for biological processes, development and metabolic processes are emphasised.

Directly annotated Gene Ontology mappings and Tissue expression locations (Fig. 21.5) further enhance the complexity of the heat stress response, as noted in Fig. 21.4 above. The response involves KEGG metabolic pathways in the liver, during foetal development, the digestive system, brain, heart, blood, and several other organs. The liver is an important regulator of growth and metabolism. It controls many physiological processes that are impacted by heat stress (Hubbard et al. 2019). These authors also show that changes in the cell membrane exert important downstream effects on heat stress response genes and metabolites (Fig. 21.5b). Other organs and structures are also important. For example, interleukins (Fig. 21.4a) are expressed in white blood cells and endothelium, while melanocytes (Fig. 21.4b) in the skin, eye, meninges and bones. The thymus (Fig. 21.5b) makes up part of the immune system and is responsible for T cell differentiation and development. Heat stress also affects the digestive and renal systems, heart and blood flow, as well as parts of the brain such as the hippocampus, cerebellum and hypothalamus.

An increase or decrease in the frequency of genetic variants in a population can be caused by natural or artificial selection pressures. Selection signatures are regions of the genome that hold functionally important sequence variants. They, therefore, are or have been under selection (natural or artificial), creating specific patterns of DNA. The reach of such signatures in the genome, up- and downstream of this functional variant, is a consequence of the so-called hitchhiking effect. Therefore, the increase in the prevalence of advantageous alleles under positive directional selection will leave distinctive signatures (patterns of genetic variation) in the DNA sequence.

Local adaptation selection signatures can be studied using whole-genome data. Spatial association with these molecular markers is also possible as candidate loci for adaptation traits can be identified through genome-environment associations (Stucki et al. 2017). Measuring Local Indicators of Spatial Association (LISA) for these candidate loci enables means that we can evaluate how similar genotypes associate spatially (Cesconeto et al. 2017). Paim et al. (2018) described the main statistical

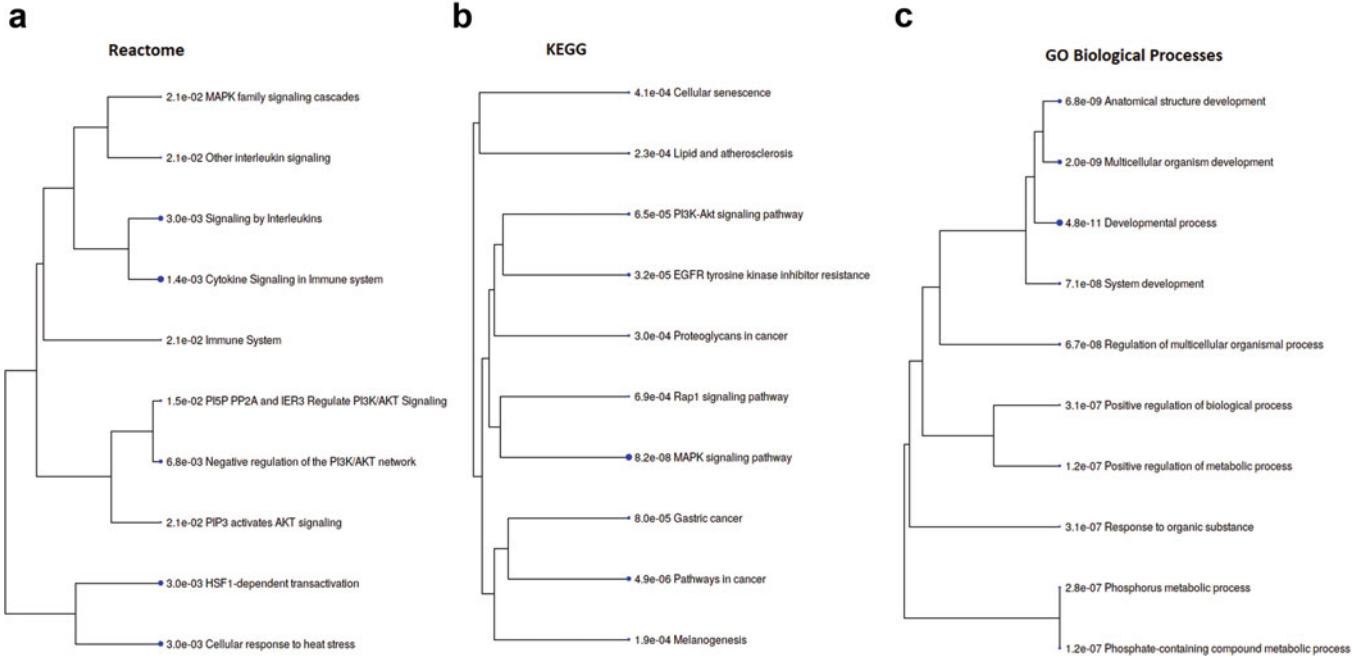

Fig. 21.4 A hierarchical clustering tree summarising the correlation among significant pathways from the Enrichment analysis using Reactome (**a**), KEGG (**b**) and Gene Ontology (GO) analyses (**c**). Pathways with many shared genes are clustered together (http://bioinformatics.sdstate.edu/go/). Numbers are False Discovery Rate (FDR). Bigger blue dots indicate more significant P-values

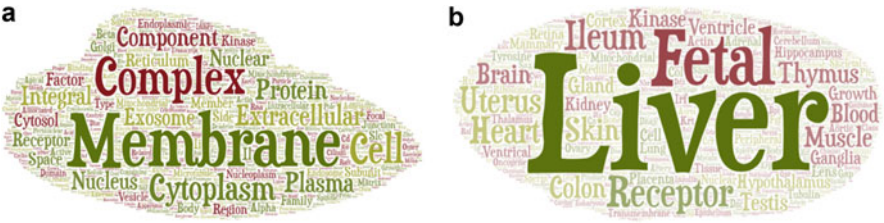

Fig. 21.5 Word clouds for Gene Ontology mappings directly annotated by the source database (**a**) and Tissue expression locations (**b**) (https://david.ncifcrf.gov/tools.jsp)

methods and software used to analyse genomic data and the identification of selection signatures.

The spatial genetic differentiation of loci can vary due to the demographic history of a species (Hoban et al. 2017). Even without selection, all have the same influence of genetic drift and migration. Possible neutral differentiation patterns need to be separated from loci under local selection. Focusing on departures from neutrality for detecting selection is difficult. Random processes affect each locus in different manners. The population structure and demography of the species in question also affect differentiation distribution. This can be problematic when there is a high average level of differentiation. The variance in F_{ST} values (Fixation index) among loci increases with average F_{ST}, even with selective neutrality, making detecting outlier loci difficult for highly differentiated populations.

The genetic bases of species adaptation to geographic conditions or climate change can be carried out using Geographical Information Systems. Manel et al. (2003) described how geographical and environmental features could facilitate genetic variation structure at population and individual levels. This method focuses on fine spatial and temporal scales, such as those with recent farm livestock migration. It has been used to identify genes responsible for adaptive evolution of species at a population level, such as quantifying the influence of spatial environment on genomic divergence and uncovering environmental factors that shape adaptive genetic variation and the genetic basis of adaptive change (Li et al. 2017). The focus in these studies needs to be on phenotypic and genetic variation so as to validate the function and adaptive generality of the detected loci. Therefore, genes involved in the regulation of metabolic pathways, as well as the adaptive phenotypes controlled by these genes, need to be identified. At the same time, Storfer et al. (2018) show that understanding the underlying demographic structure of study populations is essential when selecting genome scan methods. Species adaptability should be used to determine its distribution range, as responding to climate change depends on their landscape adaptability. This is usually determined by the genome adaptive differentiation potential as well as the species/breed's gene dispersal ability.

Learning Outcomes

- Heat stress, when combined with other stresses affects drastically the productivity of farm animals.
- Traits related to the heat stress response have low heritability values for genomic selection.
- Advanced molecular and genetic tools provide greater opportunities to identify potential biomarkers for heat stress.
- Multi-omics approaches involving genome, proteome, transcriptome, epigenome, metabolome, and microbiome could revolutionise genomic selection for heat tolerance in farm animals.

21.4 Future Perspectives

Under controlled management conditions (sufficient feeding, heat mitigation, and controlled parasite and pathogenic environment), selection for heat tolerance within highly productive breeds is likely to offer more opportunity than improving local breeds. On the other hand, crossing local and selected breeds and monitoring of heat tolerance may improve production system productivity where these conditions are not present. Research in functional genomics provides new information on HS impacts on livestock production. Genetically superior animals can be selected by identifying genes that are up- or down-regulated during HS.

Possible candidate genes have been identified in this work that could be associated with adaptation to HS. Stress causes a series of response mechanisms in animal physiology that harm the whole production chain. The identification of major genes positively correlated to heat tolerance can be used as markers in marker-assisted selection, Genomic selection, and gene editing programs. Also, other potential biomarkers on DNA and RNA, including the non-coding ones, have been proposed recently. However, there is still a gap between transforming these markers into tangible tools for breeding programs. Much research is still necessary.

Difficulties in breeding animals for resistance/tolerance to HS (Romero et al. 2015) include the lack of (1) an unambiguous definition of the stress phenotype; (2) a robust (or group of) biomarker to characterise the proposed phenotype; (3) reliable models (theoretical and quantitative) for prediction of how animals react to stressors; (4) a clear understanding of individual variability in responses to stress and transitions between acute and chronic stress. As a whole, these problems limit the ability to assess an individual's physiological status and develop techniques to reverse/control the effects of chronic stress before they become pathological. Essential for the success of breeding programs for improved HS tolerance is the availability of High-throughput phenotyping. Developing low-cost sensors and automated data collection and storage for genotyping reference populations is necessary for the efficient use of genomic breeding values and understanding the biology of the underpinning traits.

Understanding the physiological basis for adaptation requires increased characterisation with high-throughput single nucleotide polymorphism (SNP) assays or genome sequencing (Boettcher et al. 2015). Species-wide HapMap studies and multi-species studies are the first step in understanding the link between the genome and adaptation. Nevertheless, information on more breeds, geographical areas and production system environments are required to acquire a complete picture.

Metagenomics of associated microbiome can also help understand the co-adaptation of Animal Genetic Resources with other organisms in production environments. This can add information on novel biocatalysts, enzymes, genetic linkage, as well as phylogenetic and evolutionary profiles of microorganism's community function and structure and their effects on host phenotypes.

The Multi-omics approach to addressing heat tolerance and methane emissions will become important over the next decade. Examples of a multi-omics data set include some of the following: direct and indirect selection criteria, the metagenome (e.g., rumen, reproductive, and so on), mid-infrared spectral data, in addition to information from the proteome/metabolome (protein/metabolite structure and function) and functional genomic assays (e.g., methylation, transcriptomics). Causal variants can be identified when these approaches are used together with whole-genome sequencing, leading to improved responses to selection. If significant effect variants are found, these could become candidates for gene editing. In a collaborative environment, a multi-omics approach (including genome, proteome, transcriptome, epigenome, metabolome, and microbiome) could revolutionise genomic selection for traits of interest under climate change. This requires the integration of multi-omics data analysis and machine learning. Combined with the fact that mixed models have been applied to GWAS, and can reduce the number of false-positive associations, it is clear that statistical knowledge will play a significant factor in future applications of these technologies.

Genomic selection and advanced biotechnologies such as gene editing can help improve pure- and crossbreeding programmes to include traits linked to adaptation, assuming that phenotypes are available. Therefore, programs for performance recording animals in regions suffering climate change and stressful environments are needed. Integration of geographical and genetic information is also essential and adequate referencing, organisation, storage, and dissemination to stakeholders. This requires new and improved databases and information systems and the capability to link different geographical and production scales in formats that can be analysed.

Regulatory frameworks for the use of advanced technologies such as gene editing will also determine the future of their application as animal health and consumer safety has to be considered. Therefore, cooperation between all stakeholders will be needed to optimise the genetic and genomic resources to adapt livestock to climate change.

Acknowledgements Thanks are due to the *Coordenação* de Aperfeiçoamento de Pessoal de Nível Superior (CAPES) and Conselho Nacional de Desenvolvimento Científico e Tecnológico (CNPq) for financing.

Conflict of Interest The authors declare no conflict of interest.

Appendix

Table 21.1 Pathway enrichment analysis of 1703 genes (from a total of 19,606) related to heat stress in at least three species of livestock (cattle, sheep, goats or horses)

Enrichment FDR[a]	Number of genes	Pathway Genes	Fold enrichment	Pathways
GO[b] biological processes				
3.10E-07	273	1747	1.4	Response to organic substance
6.70E-08	313	2019	1.4	Regulation of multicellular organismal process
1.20E-07	331	2180	1.4	Phosphate-containing compound metabolic process
2.80E-07	331	2198	1.4	Phosphorus metabolic process
1.20E-07	381	2579	1.3	Positive regulation of metabolic process
2.00E-09	471	3208	1.3	Multicellular organism development
7.10E-08	428	2944	1.3	System development
4.80E-11	558	3841	1.3	Developmental process
6.80E-09	512	3577	1.3	Anatomical structure development
3.10E-07	556	4056	1.2	Positive regulation of biological process
GO molecular function				
1.20E-08	190	1093	1.6	ATP binding
1.20E-08	197	1142	1.6	Adenyl nucleotide binding
1.30E-08	195	1132	1.6	Adenyl ribonucleotide binding
1.30E-08	224	1348	1.5	Purine ribonucleoside triphosphate binding
1.10E-08	234	1413	1.5	Purine nucleotide binding
1.30E-08	232	1410	1.5	Ribonucleotide binding
1.10E-08	261	1609	1.5	Nucleotide binding
1.10E-08	261	1609	1.5	Nucleoside phosphate binding
1.10E-08	293	1862	1.4	Small molecule binding
1.10E-08	320	2065	1.4	Anion binding
KEGG[c]				
3.20E-05	25	76	3	EGFR tyrosine kinase inhibitor resistance
1.90E-04	26	91	2.6	Melanogenesis
8.00E-05	35	136	2.3	Gastric cancer
8.20E-08	65	263	2.2	MAPK signalling pathway
4.10E-04	33	137	2.2	Cellular senescence
2.30E-04	39	168	2.1	Lipid and atherosclerosis
3.00E-04	41	183	2	Proteoglycans in cancer
6.90E-04	40	185	2	Rap1 signalling pathway
6.50E-05	63	307	1.9	PI3K-Akt signalling pathway
4.90E-06	92	471	1.8	Pathways in cancer

(continued)

Table 21.1 (continued)

Enrichment FDR[a]	Number of genes	Pathway Genes	Fold enrichment	Pathways
Panther[d]				
4.30E-02	11	39	2.6	Interleukin signalling pathway
4.20E-02	16	64	2.3	Cadherin signalling pathway
4.30E-02	16	68	2.1	PDGF signalling pathway
2.50E-02	25	113	2	Inflammation mediated by chemokine and cytokine signalling pathway
4.30E-02	20	91	2	Angiogenesis
2.00E-02	33	157	1.9	Wnt signalling pathway
Reactome[e]				
3.00E-03	10	19	4.8	HSF1-dependent transactivation
3.00E-03	21	69	2.8	Cellular response to heat stress
6.80E-03	20	68	2.7	Negative regulation of the PI3K/AKT network
1.50E-02	19	67	2.6	PI5P PP2A and IER3 regulate PI3K/AKT signalling
2.10E-02	28	125	2	PIP3 activates AKT signalling
2.10E-02	33	156	1.9	Other interleukin signalling
2.10E-02	34	161	1.9	MAPK family signalling cascades
3.00E-03	57	285	1.8	Signalling by interleukins
1.40E-03	71	364	1.8	Cytokine signalling in immune system
2.10E-02	170	1188	1.3	Immune system
Wikipathways[f]				
5.30E-04	18	51	3.2	Thyroid Stimulating Hormone (TSH) signalling pathway
1.10E-02	14	45	2.8	Kit receptor signalling pathway
1.30E-02	14	48	2.6	Endochondral ossification
1.30E-02	14	48	2.6	Oncostatin M signalling pathway
1.20E-02	17	63	2.4	Leptin signalling pathway
9.50E-04	30	121	2.2	Integrated breast cancer pathway
9.50E-04	31	127	2.2	EGF/EGFR signalling pathway
2.10E-02	19	80	2.1	WP3231 senescence and autophagy in cancer
2.40E-04	43	182	2.1	MAPK signalling pathway
1.20E-02	25	109	2.1	BDNF signalling pathway
Cellular component				
3.50E-03	9	17	4.8	Desmosome
1.10E-07	346	2302	1.4	Cytosol
3.50E-03	373	2802	1.2	Intracellular non-membrane-bounded organelle
3.50E-03	373	2803	1.2	Non-membrane-bounded organelle
1.60E-03	465	3526	1.2	Protein-containing complex
1.50E-02	338	2571	1.2	Nucleoplasm
3.50E-03	404	3076	1.2	Membrane-enclosed lumen

(continued)

Table 21.1 (continued)

Enrichment FDR[a]	Number of genes	Pathway Genes	Fold enrichment	Pathways
3.50E-03	404	3076	1.2	Organelle lumen
3.50E-03	404	3076	1.2	Intracellular organelle lumen
8.90E-03	374	2856	1.2	Nuclear lumen
All available gene sets				
4.90E-08	234	1413	1.5	Purine nucleotide binding
4.30E-08	261	1609	1.5	Nucleotide binding
4.30E-08	261	1609	1.5	Nucleoside phosphate binding
4.90E-08	293	1862	1.4	Small molecule binding
4.90E-08	313	2019	1.4	Regulation of multicellular organismal process
4.30E-08	320	2065	1.4	Anion binding
3.20E-09	471	3208	1.3	Multicellular organism development
5.30E-08	428	2944	1.3	System development
7.90E-11	558	3841	1.3	Developmental process
1.10E-08	512	3577	1.3	Anatomical structure development

[a]*FDR* false discovery rate
[b]*GO* gene ontology
[c]*KEGG* Kyoto encyclopaedia of genes and genomes—https://www.genome.jp/kegg/pathway.html
[d]*Panther* protein analysis through evolutionary relationships—http://www.pantherdb.org/pathway/
[e]Reactome-database of reactions, pathways and biological processes—https://reactome.org/
[f]https://www.wikipathways.org/index.php/WikiPathways

References

Boettcher PJ, Hoffmann I, Baumung R, Drucker AG, McManus C, Berg P, Stella A, Nilsen LB, Moran D, Naves M, Thompson MC (2015) Genetic resources and genomics for adaptation of livestock to climate change. Front Genet 5:461. https://doi.org/10.3389/fgene.2014.00461

Cesconeto RJ, Joost S, McManus CM, Paiva SR, Cobuci JA, Braccini J (2017) Landscape genomic approach to detect selection signatures in locally adapted Brazilian swine genetic groups. Ecol Evol 7(22):9544–9556

Collier RJ, Baumgard LH, Zimbelman RB, Xiao Y (2019) Heat stress: physiology of acclimation and adaptation. Anim Front 9(1):12–19

Dikmen S, Khan FA, Huson HJ, Sonstegard TS, Moss JI, Dahl GE, Hansen PJ (2014) The SLICK hair locus derived from Senepol cattle confers thermotolerance to intensively managed lactating Holstein cows. J Dairy Sci 9:5508–5520

Gaughan JB, Mader TL, Holt SM, Hahn GLD, Young BA (2002) Review of current assessment of cattle and microclimate during periods of high heat load. Anim Product Aust 24:77–80

Hayes BJ, Bowman PJ, Chamberlain AJ, Goddard ME (2009) Invited review: genomic selection in dairy cattle: progress and challenges. J Dairy Sci 92(2):433–443

Hayes BJ, Lewin HA, Goddard ME (2013) The future of livestock breeding: genomic selection for efficiency, reduced emissions intensity, and adaptation. Trends Genet 29(4):206–214

Hoban S, Kelley JL, Lotterhos KE, Antolin MF, Bradburd G, Lowry DB, Poss ML, Reed LK, Storfer A, Whitlock MC (2017) Finding the genomic basis of local adaptation: pitfalls, practical solutions, and future directions. Am Nat 188(4):379–397

Hubbard AH, Zhang X, Jastrebski S, Singh A, Schmidt C (2019) Understanding the liver under heat stress with statistical learning: an integrated metabolomics and transcriptomics computational approach. BMC Genomics 20:502. https://doi.org/10.1186/s12864-019-5823-x

Li Y, Zhang XX, Mao RL, Yang J, Miao CY, Li Z, Qiu YX (2017) Ten years of landscape genomics: challenges and opportunities. Front Plant Sci 8:2136

Manel S, Schwartz MK, Luikart G, Taberlet P (2003) Landscape genetics: combining landscape ecology and population genetics. Trends Ecol Evolut 18(4):189–197

McManus CM, Faria DA, Lucci CM, Louvandini H, Pereira SA, Paiva SR (2020) Heat stress effects on sheep: are hair sheep more heat resistant? Theriogenology 155:157–167

Olson TA, Lucena C, Chase CC Jr, Hammond AC (2003) Evidence of a major gene influencing hair length and heat tolerance in Bos taurus cattle. J Anim Sci 81(1):80–90

Paim TDP, Ianella P, Paiva SR, Caetano AR, Pimentel CMM (2018) Detection and evaluation of selection signatures in sheep. Pesq Agrop Brasileira 53:527–539

Pryce JE, Haile-Mariam M (2020) Symposium review: genomic selection for reducing environmental impact and adapting to climate change. J Dairy Sci 103(6):5366–5375

Romero LM, Platts SH, Schoech SJ, Wada H, Crespi E, Martin LB, Buck CL (2015) Understanding stress in the healthy animal–potential paths for progress. Stress 18(5):491–497

Sejian V, Maurya VP, Kumar K, Naqvi SMK (2013) Effect of multiple stresses (thermal, nutritional and walking stress) on growth, physiological response, blood biochemical and endocrine responses in Malpura ewes under semi-arid tropical environment. Trop Anim Health Prod 45: 107–116

Singh P, Ali SA (2021) Impact of CRISPR-Cas9-based genome engineering in farm animals. Vet Sci 8(7):122

Storfer A, Patton A, Fraik AK (2018) Navigating the interface between landscape genetics and landscape genomics. Front Genet 9:68

Stucki S, Orozco-terWengel P, Forester BR, Duruz S, Colli L, Masembe C, Negrini R, Landguth E, Jones MR, Nextgen Consortium, Bruford MW (2017) High performance computation of landscape genomic models including local indicators of spatial association. Mol Ecol Resour 17(5):1072–1089