Masterproef industriële ingenieurswetenschappen 2015-2016

Genetic and environmental impact on NAD⁺ levels

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Introduction

Obesity and overweight are metabolic diseases caused by an increasing sedentary lifestyle and the intake of high-fat energy dense diets by people in industrial regions and more recently in developing countries. The prevalence of overweight and obesity is closely associated to the the development of metabolic disorders such as diabetes type II, cardiovascular diseases or nonalcoholic fatty liver disease (NAFLD) [3]. Sirtuins are NAD^+ -consuming enzymes regulating the metabolism by deacetylating transcription factors, histones and proteins. Research has shown that mitochondrial activity and thereby the mitochondrial function increases through modulation of the sirtuin pathway by boosting the NAD^+ levels [2]. This could be an interesting way to prevent and treat overweight and obesity.

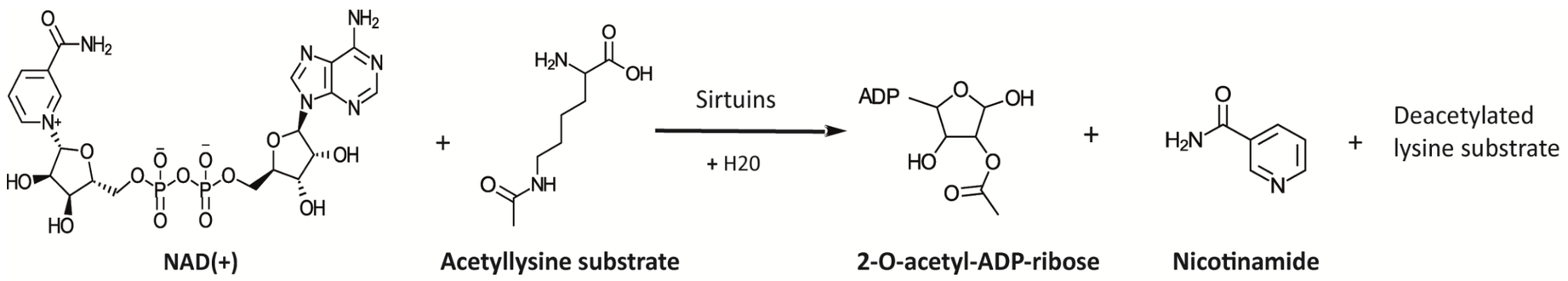

Figure 1: Sirtuins cleave NAD(+) into nicotinamide and an ADP-ribose group (adapted from [4].

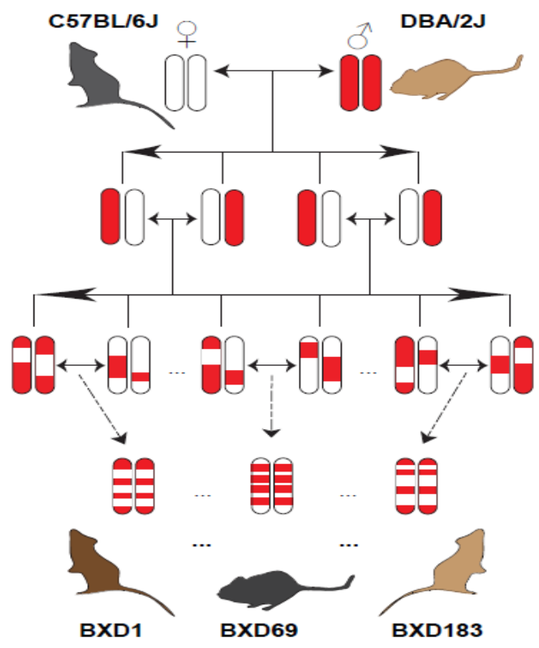

Figure 2: the BXD mouse population descends from crosses between C57BL6/J mother (B) and a DBA/2J father (D) [1].

Goal

The goal of this project is to map the quantitative trait loci (QTLs) and quantitative trait genes (QTGs) responsible for the regulation of NAD^+ levels in the BXD mouse genetic reference population (GRP). This mouse family models some aspects of the human genetic diversity and is segregating for ~ 5 million common sequence variants, similar to many human populations (e.g., Icelandic) [1]. A qualitative and quantitative method for the NAD^+ extraction and LC-MS/MS measurement was developed. The validation took place *in vitro* (AML12 mouse hepatocytes) and *in vivo* in C57BL/6J mice.

Materials and method

54 BXD strains were fed during 28 weeks on a chow (CD) or high fat diet (HFD). NAD^+ is extracted from their liver using a acidic and alkaline extraction method. Quantification was performed with LC-MS/MS. The raw data was normalized to the protein amount after mathematical correction induced by the extraction and the MS measurement. The QTLs and QTGs were mapped with GeneNetwork. Correlation analysis was performed with R.

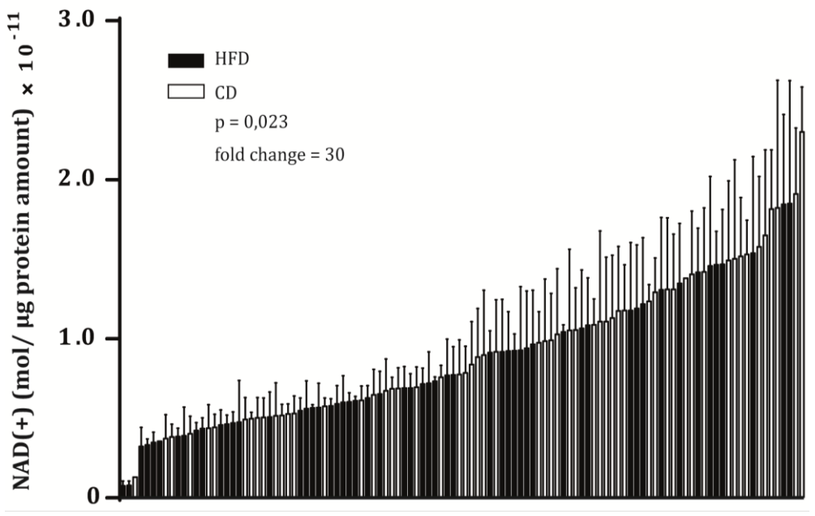

Figure 3: NAD(+) levels in liver of the BXD on HFD and CD

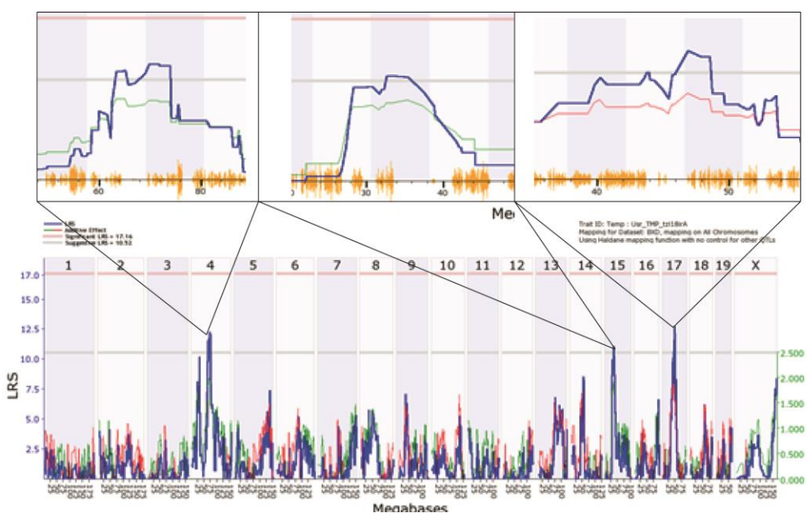

Figure 4: the suggestive QTL in strains under HFD on chromosome 4, 15 and 17.

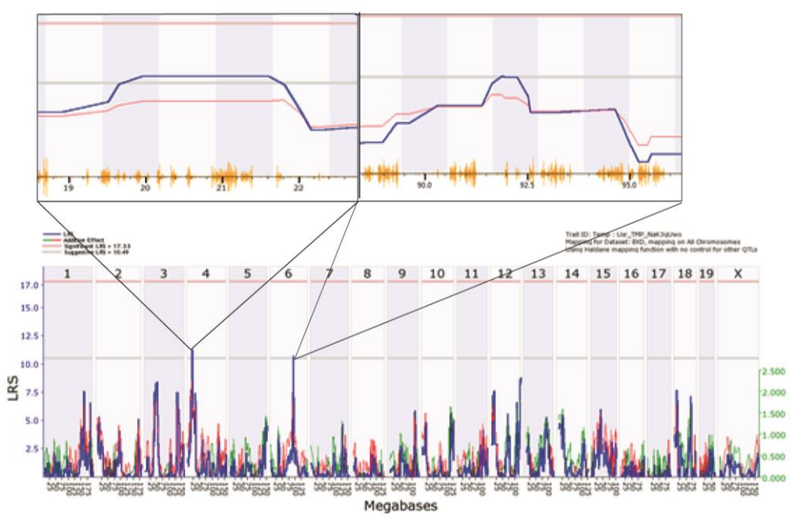

Figure 5: the suggestive QTL in strains under CD on chromosome 4 and 6.

Conclusion

We mapped 2 suggestive QTLs under CD (Figure 5) and 3 suggestive QTLs in mice fed on a HFD (Figure 4). These five suggestive QTLs combined encompass 229 QTGs with a potential role in NAD^+ regulation. Since there is no correlation observed between strains on CD and HFD, it is concluded that there is no strong genetic effect on the regulation of the NAD^+ levels. In the future the genetic and environmental factors that contribute to determining NAD^+ levels can be modeled using additional statistical tools with the data generated. A logical next step would be to detect the genetic interactions and uncover the biological pathways that are uncovered by these QTLs.

References

[1] Andreux, P. A., Williams, E. G., Koutnikova, H., Houtkooper, R. H., Champy, M. F., & Henry, H., et al. (2012). Systems genetics of metabolism: the use of the BXD murine reference panel for multiscalar integration of traits. *Cell, 150*(6), 1287-1299.
2] Houtkooper, R. H., & Auwerx, J. (2012). Exploring the therapeutic space around NAD+. *J Cell Biol, 199*(2), 205-209.
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[4]Sauve, A. A. (2010). Sirtuin chemical mechanisms. *Biochim Biophys Acta, 1804*(8), 1591-1603.

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