JOURNAL OF BASHIR INSTITUTE OF HEALTH SCIENCES

REVIEW ARTICLE

OPEN ACCESS

ARTICLE INFO
Date Received:
April 1, 2021
Date Revised:
May 2, 2021
Date Published Online
June 25, 2021

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Current trends in toxicoepigenomics research; limitations and future perspective

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ABSTRACT

Toxicoepigenomics is a newly emerged field that combines toxicity and epigenetics. It is described as heritable changes in gene activity caused by exposure to the environment and toxicants such as heavy metals without any changes to the nucleotide sequence. Epigenetics appears to play a significant role in the development of certain human diseases such as diabetes and cancer. As a result, toxicoepigenomics has emerged as a critical new area of toxicological study. Targeting DNA methylation changes as a method to identify the underlying reasons for disease development is a current trend in toxicoepigenomics research, however, this aspect of toxicoepigenomics has been thoroughly explored. Future research should consider new pathways such as microRNA (miRNA) dysregulations. Furthermore, other study approaches, such as the use of parallel *in vitro* and *in vivo* experiments, should be considered. This could be an interesting part of toxicoepigenomics research, as it will allow researchers to address a gap in current research designs.

Keywords: Toxicoepigenomics, DNA methylation, microRNA, Diabetes, Cancer

INTRODUCTION

The number of chemicals that have been reported as having "epigenetic toxicity" is rising. The Society of Toxicology (SOT) 2016 Toxico Epigenetics conference in Tysons, Virginia, USA, focused on epigenetic toxicity. Epigenetic toxicity is a process in which a chemical material alters the epigenome and has harmful effects on living organisms, which may explain the long-term consequences of toxins and disease susceptibility caused by environmental factors such as chemicals [1]. Epigenetic toxicity is a new field of research that combines toxicology with epigenetics to control gene expression and phenotypes without affecting the nucleotide sequence of the genes. Genes that are not necessary during development but are essential after growth are not affected by epigenetic changes [2].

The epigenome is a dynamic regulatory system that determines how genomic information is used to monitor how body cells, tissues, organs, and even individuals react to their environment. Toxicant exposure, food, stress, and socioeconomic position all have an impact on the epigenome, which is a fundamental regulator of gene expression [3]. To recognize the inconsistency in responsiveness to the effects of environmental toxicants, traditional toxicological models rely on factors such as an individual's age, genetic polymorphisms, and disease state. However, these factors are neither sufficient to authentically recognize an individual's differential response nor can they modify the factors that can be leveraged to dilute the effects of environmental toxicants [4]. Individuals' epigenomes, on the other hand, are altered in a variety of ways by interactions with both chemical and nonchemical

elements of the environment, making it a potential tool for public health promotion [5, 6]. Environmental variables are responsible for up to 20% of malignancies, 31% of cardiovascular problems, 42% of stroke cases, and 35% of chronic obstructive pulmonary disease cases, according to a recent analysis by a World Health Organization-convened expert group [7]. Environmental pollutants can induce or aggravate asthma, obesity, and other chronic disorders. Chronic disorders such as asthma, obesity, Alzheimer's disease, diabetes, infertility, and ovarian dysgenesis syndrome can all be started or worsened by environmental toxins. Environmental chemical pollution is predicted to cost more than ten percent of world GDP in terms of health and socioeconomic losses [8]. In the present article, we summarized the current trends in *toxicoepigenomics* research, limitations and future perspective.

CURRENT TRENDS IN TOXICOEPIGENOMICS RESEARCH

Toxicoepigenomics is a fast emerging discipline that provides new insights into the mechanisms underlying toxicant exposure-related vulnerability and diseases, but the practicality of using epigenetic data in public health and risk assessment is still unknown. As a result, it's critical to explore current trends, possible uses, and anticipated constraints that researchers are encountering when incorporating epigenetic data into human health assessment models. Toxicoepigenomics has the potential to meet critical demands in both science and applied toxicology [9].

Several research themes have evolved in recent years. The DNA methylation analysis investigations, for example, were conducted on human samples, whereas the miRNA research was primarily dominated by *in vitro* experiments. Based on current trends in toxicoepigenomics research, numerous gaps have been identified that must be filled to fully understand the molecular mechanisms behind toxicant-related epigenetic alterations [10, 11]. The consistency of epigenetic modifications happening in the experimental platform and the type of designed study such as *in vitro*, *in vivo*, and human investigations, as well as the relevance of such epigenetic alterations in changing the risk of disease in the exposed populations. It's crucial to investigate the negative consequences of environmental toxins at the systemic level, where both epigenetic and non-epigenetic modifications must be considered. The precise image of a toxicant-induced disease reveals a complicated interaction between a wide variety of signaling events taking place within the cell [12, 13].

Current literature shows that exposure to environmental toxicants mainly associated with changes in DNA methylation pattern both at gene-specific and genome-wide levels, similarly, environmental toxicants can influence histone modifications and dysregulation of miRNA which in turn affect the expression of genes [14]. However, certain important aspects are still unclear and need further investigations. Hence, it is important to explore the association between epigenetic alterations and disease development because the toxicants related epigenetic effects on the development of human diseases are more likely to rely on a variety of factors such as the genotype, exposure to a mixture of toxicants and other factors such as diet [15]. In addition, it is unclear that how the epigenetic alterations affect disease development directly, as several mechanistic associations have been established already, however it is uncertain whether there exists any linkage between epigenetic alterations and the development of disease after exposure to environmental toxicants. Still, the stability of epigenetic alterations and their utility has been put under investigation to find out the exact epigenetic mechanism because some epigenetic alterations might be adaptive and are not responsible for the development of diseases [16].

Apart from widely conducted studies that highlight the effects of the environmental toxicants on the epigenetic status of the cells and tissues, there are very limited consensuses about the exact changes occurring to the epigenome. Both hyper and hypomethylation have been constantly reported at the gene-specific and genome-wide levels [17]. The reason behind these variations in findings can be endorsed in some cases to certain technical and experimental details as environmental toxicants exhibit various physiological effects depending on concentration and time of exposure to specific toxicants [18]. Though, it seems that experiments conducted on genome-wide methylation status mostly exhibit hypomethylation predominantly as compared to hypermethylation. While on the other hand, the assays conducted on a single gene can exhibit either hypermethylation or hypomethylation pattern, mostly it depends on the type of gene, cell and tissue involved [19]. The pattern of DNA methylation suggests that environmental toxicants such as arsenic can cause both hypomethylation and hypermethylation at the same time. Though, it is clear that both hypermethylation and hypomethylation occur simultaneously but at different gene levels [20].

Tumors of various types display a range of underlying epigenetic alterations, which might be the cause of aberrant changes in the expression of certain genes involved in all cancer types. The epigenetic alterations occurring in tumor development can be

reversed by the use of small molecules which shows inhibitory effects on the action of those enzymes involved in maintaining the epigenetic state of the cell [21]. For example, certain agents such as DNA-demethylation agents have revealed significant inhibitory effects against specific hematological cancer, though the activity of such kind of inhibitory agents in terms of solid tumors remains unknown. Also, there are major challenges in the application of epigenetic therapy and maintaining a pharmacodynamics response. The use of histone lysine methyltransferases is a potential epigenetic target that needs to be explored in the future [22].

Even though there is some DNA methyltransferase (DNMT) enzymes and histone deacetylase inhibitors (HDAC), which have been already approved by the food and drug administration (FDA), but still the expansion of inhibitors that directly targeting histone methylation is in the early stages of research and need more time to fully equip this area of epigenetics for the benefits of human beings and to cure certain disorders such as cancer and diabetes [23, 24]. It is therefore important to investigate the fact that why enzymes that can fix these epigenetic marks are essential targets in cancer treatment. Hence, it has been proved that the resultant HDMTs are keenly representing the potential target class in terms of cancer treatment [25].

It is critical to enhancing the precision and efficacy of existing epigenetic treatment designs, as this will aid in recognizing epigenetic changes in the context of cancer and cancer-specific targets, as well as promoting the notion of environmental toxicants linked to epigenetic changes. Since the inter-relationships of epigenetic main regulators are complicated hence selecting compounds based on *in vitro* inhibition of a given enzyme would be a smart idea, as this will be a clear strategy for future direction [26].

LIMITATIONS IN TOXICOEPIGENOMICS STUDIES

The researchers have prioritized investigating wide verities of genes which would be represented as a key tool for the evaluation of DNA methylation status, this will create difficulties in comparing the toxic effects of the environmental chemicals on the various model systems. Similarly, the majority of the researchers choose to use a single technique to assess the DNA methylation changes, which provides a confusing conclusion. Hence, the different methods used for the determination of DNA methylation analysis can provide distinct sorts of information with different preferences. By keeping in mind such limitations in toxicoepigenomics studies and the use of proper methods for the purpose to get the chosen research results can help in fixing some of these problems occurring during the research work (Table 1). Taking the advantages of the current technologies in the area of genomics, future research will focus to relate the DNA methylation changes with transcription for the purpose to compare the desired model system on a global basis with the resolution at the sequence level [19].

The most important limitation is the absence of gene/tumor specificity. To reverse the process of silencing genes via new approaches like DNMTs and HDACs, the available compounds in the market are not enough to target gene-specific or specific cell epigenetic deregulation at the genome level. Following treatment of DNA methyltransferase inhibitors (DNMTi) or histone deacetylase inhibitors (HDACi), the subsequent hypomethylation of the DNA of gene promoters' regions may similarly also happen on the tumor silenced oncogenes. So, the availability of present data is not enough to fulfill the gap, that's why need to focus on epigenetic therapy. It is important to clarify that either the observed epigenetic changes in the animal models also occurring in the human tissues and cells. Also, It will be worthy to notice whether a triggered change in the epigenetic setting of respective cells can induce diseases or not such as alterations in the DNA methylation pattern due to exposure to environmental toxicants which end up in the development of cancer [27-29]. It is evident that symptoms, for example, allelic excitation or genomic instability could develop following a long treatment, rendering their location significantly more problematic [30]. Currently, the advancement of operators particularly focusing on inhibited keys such as TSGs is an option that could decrease the undesirable cellular toxicity and has just demonstrated some early encouraging accomplishment [31]. Another limitation of toxicoepigenomics is tissue dependence, despite the useful results obtained from the HDAC and the inhibitors of DNMT which confirms the detection of the hematological tumors. This problem is still disparately to be overcome with the help of combination of following are likely to be maintain [32]. Initially, DNMT is mostly dependent on DNA joining and also their positive effect is dependent on high proliferative rates. The cancers related to blood or other body fluids have high proliferation and cell division as compared to tissue tumors. Because of this available DNMT have poor effects on solid cancers due to low stability. Finally, hematological tumors have a low mutation which maintains apoptotic mechanisms, may add to the poor efficacy in solid malignancies [33].

FUTURE PERSPECTIVES

In the field of toxicoepigenomics, sophisticated technical advances such as methylation-sensitive PCR, next-generation sequencing, and microarrays have recently emerged as valuable tools for detecting and analyzing epigenetic alterations on a genome-wide scale. Similarly, using these methods to investigate gene expression would provide a comprehensive picture of the changes linked to gene expression. The chromatin methylation changes can be coupled with gene-expression changes using a combination of bioinformatics techniques. This will establish a connection between DNA methylation and gene expression patterns in the selected tissues [34]. Next-generation sequencing technologies will soon assist in answering questions and addressing ambiguities surrounding gene-specific versus global DNA methylation. Furthermore, other underlying epigenetic mechanisms that regulate gene expressions, such as histone modifications and mRNA, must be thoroughly investigated. The concerns about the model system used in epigenetic studies for the investigation of epigenetic changes induced by toxicants' effects are more difficult [8]. The researchers working on these issues are having difficulties deciding on toxicant exposure assessments, sample collection, and endpoints to quantify the changes. The main question from the standpoint of the environment is how close the model system is to human exposures. The application of animal model systems to human exposures during laboratory studies is unknown [35]. However, the field of toxicoepigenomics may reach a point of agreement in the coming years. For example, deciding the best current model framework for predicting the incidence of diseases linked to toxicant exposure in humans is difficult. Nevertheless, it would be a milestone in the field of toxicoepigenomics if it was understood that the same epigenetic alterations observed in animal model systems are also occurring in target human tissues [36]. The current research trends, gaps and future perspectives regarding toxicoepigenomics have been illustrated in Table 1.

Table 1: Current findings, limitations and future perspective in toxicoepigenomics research

Study	Current trends	Limitations	Future perspective
Chemical's analysis	Environmental toxicants are responsible for inducing epigenetics changes	Epigenetic alterations occur due to biofilm and microbes in chip DNMTi targets DNA globally	Testing alone the biofilm and microorganisms for toxicoepigenomics alteration
Omics and biological systems	Environmental toxicants result in a unique pattern influencing gene expression with down-regulation of stress response of the associated genes	Analysis restricted to differential expression which revealed only single time-points which is very slight validation	The application of advanced systems and biology tools such as proteomics and metabolomics and next-generation sequencing to measure the epigenetic changes
Drug interactions for cancer therapy	Drugs and several environmental toxicants exhibit synergistic effects and enhance the activity of a variety of antibiotics, DNMT and histone deacetylase inhibitors are approved drugs having limitations	Vigorous *in vivo* and *in vitro* clinical trials are essential, Isoform-specific histone deacetylase inhibitors are gradually developing which involve in pan inhibitors	Epigenetics approved drugs and environmental chemicals are needed to be evaluated for synergistic additive or antagonistic activities Determine the chemical-specific difference in response using toxicoepigenomics approaches Histone lysine methyltransferases are new targeted class because of

	specificity and cancer involve numerous enzymes

CONCLUSION

Toxicoepigenomics disorders require effective and complicated epigenetic therapeutics that will help us better comprehend the abnormal epigenetic landscape in cancers, neurological diseases, and immunological disorders. The use of *in vitro* techniques to determine the selectivity of drugs for inhibiting certain enzymes would help to realize the complexity of the interaction between epigenetic and biological systems. More research on gene expression changes in cellular circumstances through epigenetic networks at an earlier stage is essential, particularly in cancer stem cells. It is now possible to determine the underlying epigenetic alterations both at the gene-specific and genome levels, the recent advances in the field of methylation-sensitive sequencing microarray techniques are highly appreciated. The researchers would be able to address the limitations in the field of toxicoepigenomics using next-generation sequencing methods. It's also worth mentioning which animal model should be used in human physiology lab experiments. This will make it much easier to link epigenetic changes in animal models to human research. Taking into account, the aforementioned techniques could aid in the formation of a consensus in the field of toxicoepigenomics.

AUTHOR CONTRIBUTION: All the authors have directly participated in the planning or drafting of the manuscript, read and approved the final version.

CONFLICTS OF INTEREST: The authors declare no conflict of interest.

ACKNOWLEDGMENTS: This article is the outcome of an in-house financially non-supported study.

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