

Epigenetics: An Innovative Approach for Biotechnology and Food Science

Medine Gulluce, Burak Alaylar, Taha Yasin Koc, and Mehmet Karadayi

Abstract—Recently, epigenetics has been one of the most important advances in the biology and associated fields due to its valuable potential to explain how the primary DNA sequence of an organism is affected by the environmental conditions and which changes occur in the phenotype depending on related changes. Understanding rules of epigenetics and its fundamental mechanisms is essential to get a clear comprehension for many cellular processes such as quiescence, proliferation, differentiation, migration, etc., and it enables development of new technologies for various areas especially including biology, agriculture, medicine, biotechnology and food sciences. Hence, the present study was designated to review basic principles of epigenetics, recent advances in this field and its use potential for biotechnology and food applications.

Index Terms—Biotechnology, epigenetics, food science, gene expression regulations.

I. INTRODUCTION

Epigenetics, initially introduced in 1942 by Conrad Waddington, is a scientific discipline whose researches mainly focus on phenotypic alterations that become under the control of various environmental stimuli [1]–[4]. In general, the term of epigenetics refers to “above or beyond genetics” and includes heritable phenotypic changes that arise independent of primary DNA sequence [5].

Each organism, whether in a unicellular or multicellular form, has to face constantly changing environment and respond to the external impulses in each moment of its life cycle. In this manner, epigenetic mechanisms of the cell play a crucial role. Recent studies showed that these mechanisms, including DNA cytosine methylation and hydroxymethylation, and acetylation, methylation, phosphorylation, ubiquitination, SUMOylation of the lysine (K) and/or arginine (R) residues of histones, generally regulate the transcription and cause a cellular response to the stimulus. The response determines fate of the affected cell by altering expression profile of the genomic information [5]. Therefore, a clear understanding of epigenetic mechanisms enables a better conception for the link between changes in expression of the genetic information within primary DNA sequence and effects of environmental factors. As a consequence of this, it allows development of novel approaches and application strategies for various fields that consist of biological systems.

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II. BASIC MECHANISMS OF EPIGENETICS

Although there are several different classification types, the epigenetic mechanisms regulating gene expression are fundamentally divided into two subcategories as indirectly and directly acting mechanisms [6].

A. Indirectly Acting Mechanisms

This group contains post-transcriptional mechanisms, especially associated with non-coding RNAs (siRNA and miRNA), which affect mRNAs and inhibit synthesis of the related protein [6], [7].

B. Directly Acting Mechanisms

These mechanisms are also divided into two subgroups as chromatin remodeling and DNA modifications [6].

Chromatin remodeling basically consists of histone modifications such as acetylation, methylation, phosphorylation, S-nitrosylation, ubiquitination, SUMOylation, proline isomerization, ADP-ribosylation, deimination and non-covalent histone modifications [6]–[8].

Acetylation: Histone acetylation reactions are catalyzed by a specific enzyme group, called as histone acetyl transferases (HATs). In this reaction, an acetyl group from acetyl-CoA is transferred to the lysine ϵ -amino groups on the N-terminal tails of histones. This causes that the condensed chromatin is transformed into a more relaxed form and it results in a transcriptional activation. Contrary to this mechanism, deacetylation reactions are catalyzed by histone deacetylases (HDACs) and NAD⁺-dependent sirtuin family deacetylases. This reaction generally results in re-condensation of the chromatin and inhibition of the transcription [5]–[11].

Methylation: This mechanism is mainly depending on histone methyl transferases (HMTs). Lysine and arginine residues of histones are methylated by HMTs and these modifications play crucial roles in regulation of transcription (activation, elongation or repression) and DNA repair. More recent studies also pointed out that there are several enzymes responsible for demethylation of histones [5]–[12].

Phosphorylation: This type of histone modifications is catalyzed by various specific protein kinases. In the molecular mechanism, the enzyme causes the addition of a phosphate (PO₄) group to a specific part of histone molecules. For instance, Aurora-B kinase phosphorylates the serine 10 of histone H3 (H3S10) during mitosis. On the other hand, it is known that phosphatases mediate removal of the phosphate groups and reverse the reaction.

Although its exact role in the epigenetic regulations is unknown, histone phosphorylation is closely related to chromosomal condensation, transcriptional regulation and DNA repair [5]–[13].

S-nitrosylation: Protein S-nitrosylation, a type of

post-translational modification, represents the addition of an NO group to a Cys thiol to form an S-nitrosoprotein (SNO-protein). It plays an important role in cellular signaling pathways and regulation of various functional proteins [6]-[15].

Ubiquitination: In eukaryotes, ubiquitin is a small protein with 76 amino acids. One or more ubiquitin monomers covalently attach to the lysine residues of histones by ubiquitination and these are named as monoubiquitination and polyubiquitination, respectively. The related studies previously showed that monoubiquitination causes several modifications in protein functions, while polyubiquitination marks the proteins for degradation [6]-[16].

SUMOylation: This mechanism includes the addition of a small ubiquitin-related modifier protein (SUMO), approximately consists of 100 amino acids, to histones and causes a transcriptional repression [6]-[17].

Proline Isomerization: One of the recently identified histone modification mechanism is proline isomerization. It was initially reported with discovery of Frp4, a proline isomerase that accelerate switching between *cis* and *trans* conformations of prolines 30 and 38 on histone H3 tail. The conformational status of prolines regulates methylation aspects of lysine residues on histones [8].

ADP-ribosylation: Histones can be mono or poly ADP-ribosylated by ADP ribosyl transferases (ARTs) and poly (ADP-ribose) polymerases (PARPs), respectively. The former is called as mono-(ADP-ribosyl)ation and plays important roles in the cellular responses to genotoxic stress and physiological conditions (cell cycle stage, proliferation and degree of terminal differentiation). The latter, poly(ADP-ribosyl)ation (PARation), is related to the DNA repair mechanism [8].

Deimination: In this process, an arginine is converted to a citrulline by a related enzyme, for instance, arginines in H3 and H4 can be converted to citrullines by the PADI4 enzyme. This causes an antagonistic effect against arginine methylation due to the avoiding activity of citrullines from being methylation [8].

Non-covalent histon modifications: Histone modifications also involve various non-covalent mechanisms including intrachromosomal or interchromosomal interactions, histone exchanges, chromatin repair and interactions with non-coding RNAs (siRNA and miRNA) [6]-[9].

DNA methylation is the most studied and well-known type of DNA modifications. DNA methylation is catalyzed by DNA methyltransferases (DNMTs), and it, together with chromatin modifications, determines most of epigenetic programs. In its molecular mechanism, DNMTs catalyze a similar reaction with HMTs in which the enzyme transfers a methyl group from S-adenosyl methionine (SAM) to the substrate (target DNA molecule) with the formation of S-adenosyl homocysteine (SAH) as a by-product [5].

DNA methylation was formerly considered as a relatively static epigenetic sign due to the chemically stability of covalent methyl groups. However, recent studies on the cellular methylome profiles of embryonic development stages suggested the presence of enzymes responsible for the removing methyl groups. For example, the recently identified TET family enzymes hydroxylate the 5-methylcytosine of DNA [5]-[19].

In this context, several examples for modifications of histones and related enzymes are summarized in Table I and Table II, respectively.

TABLE I: SEVERAL EXAMPLES FOR MODIFICATIONS IDENTIFIED ON HISTONES [20]

Chromatin Modifications	Residues Modified	Functions Regulated
Acetylation	K-ac	Transcription, Repair, Replication, Condensation
Methylation (lysines)	K-me1, K-me2, K-me3	Transcription, Repair
Methylation (arginines)	R-me1, R-me2a, R-me2s	Transcription
Phosphorylation	S-ph, T-ph	Transcription, Repair, Condensation
Ubiquitination	K-ub	Transcription, Repair
SUMOylation	K-su	Transcription
Proline Isomerization	P-cis>P-trans	Transcription
ADP-ribosylation	R-Cit	Transcription
Deimination	E-ar	Transcription

TABLE II: SEVERAL HISTONE-MODIFYING ENZYMES [20]

Group	Enzymes	Residues Modified
Acetyltransferase	HAT1	H4 (K5, K12)
	CBP/P300	H3 (K14, K18) H4 (K5, K8) H2A (K5), H2B (K12, K15)
	PCAF/GCN5	H3 (K9, K14, K18)
	TIP60	H4 (K5, K8, K12, K16) H3 (K14)
	HB01 (ScESA1, SpMST1)	H4 (K5, K8, K12)
	ScSAS3	H3 (K14, K23)
	ScSAS2 (SpMST2)	H4K16
	ScRTT109	H3K56
Deacetylase	SirT2 (ScSir2)	H4K16
Proline Isomerase	ScFPR4	H3P30, H3P38

III. POTENTIAL APPLICATIONS IN BIOTECHNOLOGY AND FOOD SCIENCE

TABLE III: SEVERAL HISTONE-MODIFYING ENZYMES (CONTINUED)

Group	Enzymes	Residues Modified
Lysine Methyltransferase	SUV39H1	H3K9
	SUV39H2	H3K9
	G9a	H3K9
	ESET/SETDB1	H3K9
	EuHMTase/GLP	H3K9
	CLL8	H3K9
	SpCIR4	H3K9
	MLL1	H3K4
	MLL2	H3K4
	MLL3	H3K4
	MLL4	H3K4
	MLL5	H3K4
	SET1A	H3K4
	SET1B	H3K4
	ASH1	H3K4
	Sc/Sp SET1	H3K4
	SET2 (Sc/Sp SET2)	H3K36
	NSD1	H3K36
	SYMD2	H3K36
	DOT1	H3K79
	Sc/Sp DOT1	H3K79
	Pr-SET 7/8	H4K20
	SUV4 20H1	H4K20
	SUV4 20H2	H4K20
	SpSet 9	H4K20
	EZH2	H3K27
	RIZ1	H3K9
	Lysine Demethylases	LSD1BHC110
JHDM1a		H3K36
JHDM1b		H3K36
JHDM2a		H3K9
JHDM2b		H3K9
JMJD2A/JHDM3A		H3K9, H3K36
JMJD2B		H3K9
Arginine Methyltransferases	JMJD2C/GASC1	H3K9, H3K36
	JMJD2D	H3K9
Serin/Threonine Kinases	CARM1	H3 (R2, R17, R26)
	PRMT4	H4R3
	PRMT5	H3R8, H4R3
Ubiquitinases	Haspin	H3T3
	MSK1	H3S28
	MSK2	H3S28
	CKII	H4S1
	Mst1	H2BS14
Ubiquitinases	Bmi/Ring1A	H2AK119
	RNF20/RNF40	H2BK120

Many researches in the epigenetics field mainly focus on human health care issues including embryonic development, aging and formation of a broad range of diseases such as varied types of cancer, type 2 diabetes mellitus, obesity, inflammation, cardiovascular diseases and neurocognitive

disorders. Epigenetics is also thought as one of the best candidates for explanation of the unknown etiology of many human diseases. According to these frames, it is getting clear that a better understanding of epigenetic rules and related mechanisms will lead up to get more comprehensive and efficient results in the prevention and treatment strategies [21]-[23].

TABLE IV: SEVERAL EXAMPLES FOR EPIGENETIC DRUGS [21]

Target	Drug
DNA methylation	5-Azacytidine
	5-Aza-2'-deoxycytidine
	5-fluoro-2'-deoxycytidine
	Zebularine
	Procainamide
	Epigallocatechin-3-gallate
Histone deacetylase	Psammaplin A
	Antisense oligomers
	Phenylbutyric acid
	Suberoylanilide hydroxamic acid
	Depsiptide
	Valproic acid

Current status of knowledge on the epigenetic mechanisms enables development of promising strategies against many of human disorders related to phenotypic alterations occurring under environmental conditions and independent genomic sequence changes. In this manner, reversibility of epigenetic alterations is the most highlighted property, which provides a possibility to envisage the novel drug approaches in biotechnology for the future. This is called as "Epigenetic Therapy" and several examples for the drugs are presented in Table III and Table IV. Moreover, determination of epigenetic modifications on a specific gene or a region of chromatin material can be used as a diagnostic marker for the screening and prognosis of the related diseases [21]-[23].

On the other hand, epigenetics-associated studies have become exceptionally important in the nutritional field and food science because several nutrients may affect epigenetic patterns of an organism, resulting in a significant alteration in the gene expression levels, which may cause modifications in health-related phenotypes. This new field is called as nutritional epigenetics [24], [25].

Nutritional epigenetics fundamentally focuses on the alterative capabilities of nutrients and bioactive food components on the epigenetic phenomena. According to the recent advances in this field, several nutritive compounds such as folate, vitamin B-12, biotin, niacin, pantothenic acid, methionine, choline, genistein, tea catechin, resveratrol, butyrate, sulforaphane, diallyl sulfide and betaine were determined as efficient components on epigenetic mechanisms. With reference to current knowledge, any of these compounds may affect epigenetic pattern via acting two different ways. In the first proposed mechanism, a nutrient can act as a substrate for the enzymes associated with epigenetic regulation, but as an inhibitor in the second mechanism. For example, biotin is a substrate of histone biotinylation and curcumin is an inhibitor of histone acetyltransferases [24], [25].

Nevertheless, the knowledge of nutritional epigenetics is still insufficient and limited. Moreover, exact effect mechanisms of many bioactive substances also are unclear. However, the count of identified ones related to epigenetic mechanisms is drastically increasing by the development of

novel technologies and new research strategies [24], [25]. In this manner, several examples for identified active nutrients and their effect mechanisms are shown in Table V.

TABLE V: SEVERAL EXAMPLES FOR NUTRITIONAL EPIGENETICS [24]

	Nutrient or diet	Epigenetic mechanism
Embryonic Development	Folate	DNA methylation Imprinting
	Choline	DNA methylation
	Protein restriction	DNA methylation Histone modifications
	Alcohol	DNA methylation
Stem Cell	Butyrate	Histone acetylation DNA methylation
	Retinoic acid	PRC
Aging	Folate	DNA methylation
	Calorie restriction	Histone acetylation
Immune Function	Folate	DNA methylation
	Methyl-deficient diet	Histone modification MicroRNA
Cancer	Genistein	DNA methylation MicroRNA
	(-)-Epigallocatechin-3-gallate	DNA methylation PRC
	Curcumin	MicroRNA
Obesity, Insulin resistance	High-fat diet	DNA methylation MicroRNA
	Methyl-deficient diet	DNA methylation
	Curcumin	Histone acetylation
Inflammation	Resveratrol	Histone acetylation
	AdoMet	Histone methylation
	Methyl-deficient diet	MicroRNA
Neurocognition	Choline	DNA methylation Histone methylation

IV. CONCLUSION

Recently epigenetics has been one of the most promising fields in biotechnology and food science. Reversibility of its mechanisms enables improvement of new prevention and therapy strategies against several human disorders associated with epigenetic phenomenon regulated by various environmental factors.

Among all the external stimuli that cause epigenetic changes, nutrition has been the most attractive one due to its huge place in the human lifetime. Up to date, its role in embryonic development, stem cell differentiation, aging,

immune response and formation of several human diseases has been intensively studied. The count of related studies has been also increasing day by day.

In a conclusion, epigenetics will be an essential and inevitable in the near future. Its potential in development of health-related applications is clear. As a consequence, epigenetics can be thought as an innovative approach for biotechnology and food science.

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