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A review on Microarray technology in molecular diagnostics

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Abstract
Advances in technology opened many doors, increased opportunities in various research fields, and helped create tools and devices required for many analyses. One of them is microarray technology. It is widely used in numerous research areas including infection biology, detection of small noncoding RNAs, microarray-based gene expression profiling, and many more. Microarray implementation in molecular diagnostics and their use as diagnostic devices are considered a step closer to precision medicine. Further advances in microarray technology can potentially help with the identification of new pathogens, improvement in medicine, and better drug delivery.

Keywords: Microarrays, Fabrication, Molecular diagnostics, Probes

1. Introduction

Advances in technology opened many doors, increased opportunities in various research fields, and helped create tools and devices required for certain analysis. One of them is microarray technology. It's providing assistance in disease treatment, cancer research and pharmacological approach [1]. Microarray technology is based on measuring concentrations of large amount of samples, usually DNA, on small surface [2], [3]. They are widely used in numerous research areas including infection biology, detection of small noncoding RNAs, microarray-based gene expression profiling etc. [4]. With microarray technology it's possible to systematically scan whole genome and find new genetic traits and disorders that are influencing gene expressions [5], [6]. Based on sample type, there are three categories of microarrays: DNA microarrays [7], RNA microarrays [8] and protein microarrays [9]. However, DNA microarrays are most popular and widely used in different applications [2]. Molecular diagnostics on the other hand, is process used for molecule analysis in order to identify or prevent diseases [10]. Microarray implementations in molecular diagnostics and their use as diagnostic devices are considered a step closer towards precision medicine [4], [11].

2. Microarray fabrication

There are three main fabrication methods for microarrays including light-directed synthesis, robot spotting and piezoelectric ink-jet printing [12]–[14]. For light-directed synthesis, often called photolithography, either digital micro mirror devices or premade masks are used. Micro mirror devices navigate light required for every spot on microarray, thus significantly reducing costs, while on the other hand, for every nucleotide in growing strand on the microarray support, premade masks are applied, making this type of technology much more expensive [4], [12]. Robot spotting technology for microarray fabrication is one of most widely used technologies, especially in research field. Samples including oligonucleotides, PCR products or proteins are spotted and printed by either solid or split pins [4]. By piezoelectric ink-jet printing technology, oligonucleotides and cDNAs are produced. While cDNA is directly spotted, oligonucleotides require

repetitive base-by-base print layers [4].

3. Microarrays in Infection biology

Microarrays proved to be reliable technology for detection and identification of pathogenic organisms because of its sensitivity, cost, processing time and specificity [15]. There are different types of approaches for detection of pathogenic organisms including ViroChip, resequencing pathogen microarrays, universal detection arrays, GreeneChip and Lawrence Livermore microbial detection arrays [15].

3.1. ViroChip

First produced microarray for pathogen detection was ViroChip [16]. It contained 1600 probes from 140 viral genomes obtained from GenBank available at that time [15]. Working principle of ViroChip is based on spotting oligonucleotides that are synthesized on glass slide. Additionally, ViroChip can be used for identification of new viruses from the same family because of the probe design (containing conserved regions) [15].

3.2. Resequencing Pathogen microarrays

Resequencing pathogen microarrays (RPM) are composed of short probes with selected genes for target species of pathogen [17]. For each location of target gene four probes are designed including one with perfect base match and others on alternative bases [17]. The main advantage of RPM array is highly specificity for identification of strain-level target organisms. However, there are few disadvantages as well, including limited range of organisms on single array that can be covered and inability to detect novel organisms [15].

3.3. Universal Detection array

Universal detection array is based on universally applicable set of probes containing more than 14 000 probes with randomly produced sequences [18]. Based on DNA hybridization patterns various organisms can be distinguished [18]. Huge advantage of this approach is that probes can be non-specific to genomes, meaning that there is no need to change array design and update genomes. On the hand, one of the disadvantages is that signatures for known sequence organisms must be experimentally obtained for every organism of interest [15].

3.4. GreeneChip

GreeneChip is designed in order to promote unbiased, rapid and differential diagnosis of different pathogenic organisms [19]. This type of array is based on selecting certain conserved region together with few variable regions [19]. Probes are selected in a way that every pathogen is represented with at least one probe [15]. GreeneChip array proved to be more accurate with virus probes compared to bacterial ones [15].

3.5. Lawrence Livermore Microbial detection array

Initial version of Lawrence Livermore microbial detection array or shortly LLMDA was composed of target probes for all viruses and bacteria (pathogenic and non-pathogenic) [15]. This approach allows us to screen and examine each microbial genome with significant number of different probes [15]. LLMDA array showed to be highly specific and sensitive for large number of previously characterized bacterial and viral cultures. Broad coverage of viruses and bacteria is one of the important advantages of LLMDA. However, coast of probe design is main disadvantage [15].

3.6. Detection of Small Non-coding RNAs

Since small non-coding RNAs are proved to be important regulators of development, understanding their function and expanding their application is crucial [20]. Using them, especially microRNAs (miRNAs), as diagnostic markers could provide better understanding of various types of diseases including cardiovascular diseases [21], neurodegenerative diseases [22], infectious diseases [23] and cancer [24]. However, microRNA detection is challenging procedure because of its short and complex form. Accurate and sensitive microarray

assay called Agilent Technology is used for profiling of miRNAs [25]. Working principle of this assay is that nonfractioned RNA is labeled by ligation to 3' end of RNA, further interacting with 5' end of probe and contributing to the stability of complex [25]. Additionally, specific probe design allows highly specific, precise and accurate detection of closely related miRNAs [25].

4. Microarray based gene expression profiling

This type of approach is used to analyze gene transcription in order to classify diseases, understand the mechanisms related to diseases processes, identify diagnostic biomarkers and monitor the therapy response [26]. Microarray-based gene expression profiling is mainly used in research related to gene expression, since Human Genome Project revealed important knowledge of human DNA organization, and may provide and reveal important information regarding molecular mechanisms of diseases [27]. It's based on identification of clones and isolation of high-quality RNA from tissue [27]. This microarray approach is an example of how advances in technology may successfully contribute and support clinical decision-making, prediction and classification of different cancer types [28].

5. Commercial microarrays in diagnostics

Generally, microarrays used in molecular diagnostics are rare because of their sensitivity, contamination risk and reproducibility. However, generally used ones are Amplichip, MammaPrint, ViroChip etc. [4]. Amplichip is pharmacokinetic test based on microarray technology and used for clinical evaluation of treatment selection for an individual [29]. MammaPrint technology is used for breast cancer analysis, more precisely to evaluate and predict the risk of metastasis formation [30].

6. Conclusion

Microarray technology enabled researchers to monitor and analyze gene expression levels and wide range of molecules. That information can be used in medicine for numerous applications including clinical outcome prediction, therapy response and discovery of new diseases or subclasses of diseases. Although this technology has great potential, it is still not routinely used in molecular diagnostics. Main reason for that are complexity and sensitivity. Additionally, microarray procedures often require manual steps thus increasing the possibility of error. Further advances in microarray technology can potentially help with identification of new pathogens, improvement of precision medicine, better drug delivery etc.

7. References

[1] K. Kaliyappan, M. Palanisamy, R. Govindarajan, and J. Duraiyan, "Microarray and its applications," *J. Pharm. Bioallied Sci.*, vol. 4, no. 6, p. 310, 2012, doi: 10.4103/0975-7406.100283.

[2] R. Bumgarner, "Overview of DNA Microarrays: Types, Applications, and Their Future," *Curr. Protoc. Mol. Biol.*, vol. 101, no. 1, Jan. 2013, doi: 10.1002/0471142727.mb2201s101.

[3] M. B. Miller and Y.-W. Tang, "Basic Concepts of Microarrays and Potential Applications in Clinical Microbiology," *Clin. Microbiol. Rev.*, vol. 22, no. 4, pp. 611–633, Oct. 2009, doi: 10.1128/CMR.00019-09.

[4] K. Lemuth and S. Rupp, "Microarrays as Research Tools and Diagnostic Devices," in *RNA and DNA Diagnostics*, V. A. Erdmann, S. Jurga, and J. Barciszewski, Eds. Cham: Springer International Publishing, 2015, pp. 259–280. doi: 10.1007/978-3-319-17305-4_13.

[5] R. Plomin and L. C. Schalkwyk, "Microarrays," *Dev. Sci.*, vol. 10, no. 1, pp. 19–23, Jan. 2007, doi: 10.1111/j.1467-7687.2007.00558.x.

[6] P. O. Brown and D. Botstein, "Exploring the new world of the genome with DNA microarrays," *Nat. Genet.*, vol. 21, no. S1, pp. 33–37, Jan. 1999, doi: 10.1038/4462.

[7] U. Bilitewski, "DNA Microarrays: An Introduction to the Technology," in *Microchip Methods in Diagnostics*, vol. 509, U. Bilitewski, Ed. Totowa, NJ: Humana Press, 2009, pp. 1–14. doi: 10.1007/978-1-59745-372-1_1.

[8] S. C. Sealfon and T. T. Chu, "RNA and DNA Microarrays," in *Biological Microarrays*, vol. 671, A. Khademhosseini, K.-Y. Suh, and M. Zourob, Eds. Totowa, NJ: Humana Press, 2011, pp. 3–34. doi: 10.1007/978-1-59745-551-0_1.

[9] G. MacBeath, "Protein microarrays and proteomics," *Nat. Genet.*, vol. 32, no. S4, pp. 526–532, Dec. 2002, doi: 10.1038/ng1037.

[10] H. Choe, C. A. Deirmengian, N. J. Hickok, T. N. Morrison, and R. S. Tuan, "Molecular Diagnostics:," *J. Am. Acad. Orthop. Surg.*, vol. 23, pp. S26–S31, Apr. 2015, doi: 10.5435/JAAOS-D-14-00409.

[11] S. Dwivedi *et al.*, "Diseases and Molecular Diagnostics: A Step Closer to Precision Medicine," *Indian J. Clin. Biochem.*, vol. 32, no. 4, pp. 374–398, Oct. 2017, doi: 10.1007/s12291-017-0688-8.

[12] F. Yang *et al.*, "Light-Directed Synthesis of High-Density Peptide Nucleic Acid Microarrays," *ACS Comb. Sci.*, vol. 17, no. 10, pp. 608–614, Oct. 2015, doi: 10.1021/acscombsci.5b00074.

[13] O. Rando, "Printing Microarrays," *Cold Spring Harb. Protoc.*, vol. 2019, no. 9, p. pdb.prot096396, Sep. 2019, doi: 10.1101/pdb.prot096396.

[14] S. Fujita, R. Onuki-Nagasaki, J. Fukuda, J. Enomoto, S. Yamaguchi, and M. Miyake, "Development of super-dense transfected cell microarrays generated by piezoelectric inkjet printing," *Lab Chip*, vol. 13, no. 1, pp. 77–80, 2013, doi: 10.1039/C2LC40709D.

[15] K. S. McLoughlin, "Microarrays for Pathogen Detection and Analysis," *Brief. Funct. Genomics*, vol. 10, no. 6, pp. 342–353, Nov. 2011, doi: 10.1093/bfgp/elr027.

[16] D. Wang *et al.*, "Microarray-based detection and genotyping of viral pathogens," *Proc. Natl. Acad. Sci.*, vol. 99, no. 24, pp. 15687–15692, Nov. 2002, doi: 10.1073/pnas.242579699.

[17] B. Lin *et al.*, "Broad-spectrum respiratory tract pathogen identification using resequencing DNA microarrays," *Genome Res.*, vol. 16, no. 4, pp. 527–535, Apr. 2006, doi: 10.1101/gr.4337206.

[18] Y. Y. Belosludtsev *et al.*, "Organism identification using a genome sequence-independent universal microarray probe set," *BioTechniques*, vol. 37, no. 4, pp. 654–660, Oct. 2004, doi: 10.2144/04374RR02.

[19] G. Palacios *et al.*, "Panmicrobial Oligonucleotide Array for Diagnosis of Infectious Diseases," *Emerg. Infect. Dis.*, vol. 13, no. 1, pp. 73–81, Jan. 2007, doi: 10.3201/eid1301.060837.

[20] N. Bushati and S. M. Cohen, "microRNA Functions," *Annu. Rev. Cell Dev. Biol.*, vol. 23, no. 1, pp. 175–205, Nov. 2007, doi: 10.1146/annurev.cellbio.23.090506.123406.

[21] K. L. Ellis, V. A. Cameron, R. W. Troughton, C. M. Frampton, L. J. Ellmers, and A. M. Richards, "Circulating microRNAs as candidate markers to distinguish heart failure in breathless patients," *Eur. J. Heart Fail.*, vol. 15, no. 10, pp. 1138–1147, Oct. 2013, doi: 10.1093/eurjhf/hft078.

[22] R. Gandhi *et al.*, "Circulating MicroRNAs as biomarkers for disease staging in multiple sclerosis: Circulating MicroRNAs in MS," *Ann. Neurol.*, vol. 73, no. 6, pp. 729–740, Jun. 2013, doi: 10.1002/ana.23880.

[23] S. Shrivastava, "Hepatitis C virus infection, microRNA and liver disease progression," *World J. Hepatol.*, vol. 5, no. 9, p. 479, 2013, doi: 10.4254/wjh.v5.i9.479.

[24] M. D. Jansson and A. H. Lund, "MicroRNA and cancer," *Mol. Oncol.*, vol. 6, no. 6, pp. 590–610, Dec. 2012, doi: 10.1016/j.molonc.2012.09.006.

[25] P. N. D'Andrade and S. Fulmer-Smentek, "Agilent MicroRNA Microarray Profiling System," in *Next-Generation MicroRNA Expression Profiling Technology*, vol. 822, J.-B. Fan, Ed. Totowa, NJ: Humana Press, 2012, pp. 85–102. doi: 10.1007/978-1-61779-427-8_6.

[26] A. L. Tarca, R. Romero, and S. Draghici, "Analysis of microarray experiments of gene expression profiling," *Am. J. Obstet. Gynecol.*, vol. 195, no. 2, pp. 373–388, Aug. 2006, doi: 10.1016/j.ajog.2006.07.001.

[27] A. T. Weeraratna, J. E. Nagel, V. de Mello-Coelho, and D. D. Taub, "Gene Expression Profiling: From Microarrays to Medicine," *J. Clin. Immunol.*, vol. 24, no. 3, pp. 213–224, May 2004, doi: 10.1023/B:JOCI.0000025443.44833.1d.

[28] P.-E. Colombo, F. Milanezi, B. Weigelt, and J. S. Reis-Filho, "Microarrays in the 2010s: the contribution of microarray-based gene expression profiling to breast cancer classification, prognostication and prediction," *Breast Cancer Res.*, vol. 13, no. 3, p. 212, Jun. 2011, doi: 10.1186/bcr2890.

[29] K. K. Jain, "Applications of AmpliChip CYP450:," *Mol. Diagn.*, vol. 9, no. 3, pp. 119–127, 2005, doi: 10.2165/00066982-200509030-00002.

[30] B. S. Wittner *et al.*, "Analysis of the MammaPrint Breast Cancer Assay in a Predominantly Postmenopausal Cohort," *Clin. Cancer Res.*, vol. 14, no. 10, pp. 2988–2993, May 2008, doi: 10.1158/1078-0432.CCR-07-4723.