

## Appendix

### References

1. Collins, F. S., Guyer, M. S., and Charkravarti, A. (1997). "Variations on a theme: Cataloging human DNA sequence variation." *Science*, 278, 1580-1581.
2. Schafer, A. J., and Hawkins, J. R. (1998). "DNA variation and the future of human genetics." *Nature Biotechnology*, 16, 33-39.
3. Cargill, M., Altshuler, D., Ireland, J., Sklar, P., Ardlie, K., Patil, N., Lane, C. R., Lim, E. P., Kalyanaraman, N., Nemesh, J. et al. (1999). "Characterization of single nucleotide polymorphisms in coding ferions of human genes." *Nature Genetics*, 22, 231-238.
4. Halushka, M. K., Fan, J.-B., Bentley, K., Hsie, L., Shen, N., Weder, A., Cooper, R., Lipshutz, R., and Chakravarti, A. (1999). "Patterns of single-nucleotide polymorphisms in candidate genes for blood pressure homeostasis." *Nature Genetics*, 22, 239-247.
5. Cooper, D. N., Smith, B. A., Cooke, H. J., Niemann, S., and Schmidtke, J. (1985). "An estimate of unique DNA sequence heterozygosity in the human genome." *Hum. Genet.*, 69, 201-205.
6. Brookes, A. J. (1999). "The essence of SNPs." *Gene*, 234, 177-186.
7. Kruglyak, L. (1997). "The use a genetic map of biallelic markers in linkage studies." *Nature Genetics*, 17, 21-24.
8. Kruglyak, L. (1999). "Prospects for whole-genome linkage disequilibrium mapping of common disease genes." *Nature Genetics*, 22, 139-144.
9. Lander, E. S. (1996). "The new genomics : Global views of biology." *Science*, 274, 536-539.
10. Weeks, D. E., and Lathrop, G. M. (1995). "Polygenic disease: methods for mapping complex disease traits." *Trends Genet.*, 11, 513-519.
11. Lander, E. S., and Schork, N. J. (1994). "Genetic dissection of complex traits." *Science*, 265, 2037-2048.

12. NIH Guide: Methods for Discovering and Scoring Single Nucleotide Polymorphisms, [http://www.nih.gov/grants/guide\\_rfa-files/RFA-HG-98-001.htm](http://www.nih.gov/grants/guide_rfa-files/RFA-HG-98-001.htm), pp. 1-12 (Mai 2000)
13. Risch, N., and Merikangas, K. (1996). "The future of genetic studies of complex human diseases." *Science*, 273, 1516-1517.
14. Kruglyak, L., and Lander, E. S. (1995). "High-resolution Genetic Mapping of Complex Traits." *Am. J. Hum. Genet.*, 56, 1212-1223.
15. Risch, N. J. (2000). "Searching for genetic determinants in the new millennium." *Nature*, 405, 847-856.
16. Zhao, L. P., Aragaki, C., Hsu, L., and Quiaoit, F. (1998). "Mapping of complex traits by single-nucleotide polymorphisms." *Am. J. Hum. Genet.*, 63, 225-240.
17. Weiss, K. M., and Terwilliger, J. D. (2000). "How many diseases does it take to map a gene with SNPs?" *Nature Genetics*, 26, 151-157.
18. Hugot, J. P., Chamaillard, M., Zouali, H., Lesage, S., Cezard, J. P., Belaiche, J., Almer, S., Tysk, C., O'Morain, C. A., Gassull, M. et al. (2001). "Association of NOD2 leucine-rich repeat variants with susceptibility to Crohn's disease." *Nature*, 411, 599-603.
19. Ogura, Y., Bonen, D. K., Inohara, N., Nicolae, D. L., Chen, F. F., Ramos, R., Britton, H., Moran, T., Karaliuskas, R., Duerr, R. H. et al. (2001). "A frameshift mutation in NOD2 associated with susceptibility to Crohn's disease." *Nature*, 411, 603-606.
20. Sachidanandam, R., Weissman, D., Schmidt, S. C., Kakol, J. M., Stein, L. D., Marth, G., Sherry, S., Mullikin, J. C., Mortimore, B. J., Willey, D. L. et al. (2001). "A map of human genome sequence variation containing 1.42 million single nucleotide polymorphisms." *Nature*, 409, 928-933.
21. Venter, J. C., Adams, M. D., Myers, E. W., Li, P. W., Mural, R. J., Sutton, G. G., Smith, H. O., Yandell, M., Evans, C. A., Holt, R. et al. (2001). "The sequence of the human genome." *Science*, 291, 1304-51.

22. Evans, W. E., and Relling, M. V. (1999). "Pharmacogenetics : Translating functional genomics into rational therapeutics." *Science*, 286, 487-491.
23. McCarthy, J. J., and Hilfiker, R. (2000). "The use of single-nucleotide polymorphism maps in pharmacogenomics." *Nature Biotechnology*, 18, 505-508.
24. Brinkmann, U. (2001). "Polymorphisms of the Drug Transporter MDR1 and Cyp3A Enzymes." *Abstracts of the Cambridge Healthtech Institute's Second Annual Pharmacogenomics Europe Conference, Munich, Germany 23-24. Mai*
25. Landegren, U., Kaiser, R., Caskey, C. T., and Hood, L. (1988). "DNA diagnostics - molecular techniques and automation." *Science*, 242, 229-237.
26. Meldrum, D. (2000). "Automation for genomics, part one: preparation for sequencing." *Genome Res.*, 10, 1081-1092.
27. Meldrum, D. (2000). "Automation for genomics, part two: sequencers, microarrays, and future trends." *Genome Res.*, 10, 1288-1303.
28. Gut, I. G. (2001). "Automation in genotyping of single nucleotide polymorphisms." *Hum. Mutat.*, 17, 475-492.
29. Chicurel, M. (2001). "Faster, better, cheaper genotyping." *Nature*, 412, 580-582.
30. Lander, E. S., Linton, L. M., Birren, B., Nusbaum, C., Zody, M. C., Baldwin, J., Devon, K., Dewar, K., Doyle, M., FitzHugh, W. et al. (2001). "Initial sequencing and analysis of the human genome. International Human Genome Sequencing Consortium." *Nature*, 409, 860-921.
31. Sanger, F., Nickens, S., and Coulson, A. R. (1977). "DNA sequencing with chain-terminating inhibitors." *Proc. Natl. Acad. Sci. USA*, 74, 5463-5467.
32. Smith, L. M., Sanders, J. Z., Kaiser, R. J., Hughes, P., Dodd, C., Connell, C. R., Heiner, C., Kent, S. B., and Hood, L. E. (1986). "Fluorescence detection in automated DNA sequence analysis." *Nature*, 321, 674-679.
33. Cohen, A. S., Najarian, D. R., Paulus, A., Guttman, A., Smith, J. A., and Karger, B. L. (1988). "Rapid separation and purification of oligonucleotides by high-performance capillary gel electrophoresis." *Proc. Natl. Acad. Sci. USA*, 85, 9660-9663.

34. Ronaghi, M., Nygren, M., Lundeberg, J., and Nyrén, P. (1999). "Analyses of Secondary Structures in DNA by Pyrosequencing." *Anal. Biochem.*, 267, 65-71.
35. <http://www.appliedbiosystems.com>
36. Ronaghi, M., Uhlén, M., and Nyrén, P. (1998). "A Sequencing Method Based on Real-Time Pyrophosphate." *Science*, 281, 363-365.
37. <http://www.pyrosequencing.com>
38. Lander, E. S. (1999). "Array of hope." *Nature Genetics*, 21 (Suppl.), 3-4.
39. Wang, D. G., Fan, J.-B., Siao, C.-J., Berno, A., Young, P., Sapolsky, R., Ghandour, G., Perkins, N., Winchester, E., Spencer, J. et al. (1998). "Large-scale identification, mapping, and genotyping of single nucleotide polymorphisms in the human genome." *Science*, 280, 1077-1082.
40. Edman, C. F., Raymond, D. E., Wu, D. J., Tu, E., Sosnowski, R. G., Butler, W. F., Nerenberg, M., and Heller, M. J. (1997). "Electric field directed nucleic acid hybridization on microchips." *Nucleic Acids Res.*, 25, 4907-4914.
41. Sosnowski, R. G., Tu, E., Butler, W. F., O'Connell, J. P., and Heller, M. J. (1997). "Rapid determination of single base mismatch mutations in DNA hybrids by direct electric field control." *Proc. Natl. Acad. Sci. USA*, 94, 1119-1123.
42. Pastinen, T., Kurg, A., Metspalu, A., Peltonen, L., and Syvanen, A. C. (1997). "Minisequencing: a specific tool for DNA analysis and diagnostics on oligonucleotide arrays." *Genome Res.*, 7, 606-614.
43. Pastinen, T., Raitio, M., Lindroos, K., Tainola, P., Peltonen, L., and Syvanen, A. C. (2000). "A system for specific, high-throughput genotyping by allele-specific primer extension on microarrays." *Genome Res.*, 10, 1031-1042.
44. Fan, J. B., Chen, X., Halushka, M. K., Berno, A., Huang, X., Ryder, T., Lipshutz, R. J., Lockhart, D. J., and Chakravarti, A. (2000). "Parallel genotyping of human SNPs using generic high-density oligonucleotide tag arrays." *Genome Res.*, 10, 853-860.
45. Gerry, N. P., Witowski, N. E., Day, J., Hammer, R. P., Barany, G., and Barany, F. (1999). "Universal DNA microarray method for multiplex detection of low abundance point mutations." *J. Mol. Biol.*, 292, 251-262.

46. Hirschhorn, J. N., Sklar, P., Lindblad-Toh, K., Lim, Y. M., Ruiz-Gutierrez, M., Bolk, S., Langhorst, B., Schaffner, S., Winchester, E., and Lander, E. S. (2000). "SBE-TAGS: an array-based method for efficient single-nucleotide polymorphism genotyping." *Proc. Natl. Acad. Sci. USA*, 97, 12164-12169.
47. Chen, J., Iannone, M. A., Li, M. S., Taylor, J. D., Rivers, P., Nelsen, A. J., Slentz-Kesler, K. A., Roses, A., and Weiner, M. P. (2000). "A microsphere-based assay for multiplexed single nucleotide polymorphism analysis using single base chain extension." *Genome Res.*, 10, 549-557.
48. Iannone, M. A., Taylor, J. D., Chen, J., Li, M. S., Rivers, P., Slentz-Kesler, K. A., and Weiner, M. P. (2000). "Multiplexed single nucleotide polymorphism genotyping by oligonucleotide ligation and flow cytometry." *Cytometry*, 39, 131-140.
49. <http://www.luminex.com>
50. Chen, X., Zehnbauser, B., Gnirke, A., and Kwok, P. Y. (1997). "Fluorescence energy transfer detection as a homogeneous DNA diagnostic method." *Proc. Natl. Acad. Sci. USA*, 94, 10756-10761.
51. Holland, P. M., Abramson, R. D., Watson, R., and Gelfand, D. H. (1991). "Detection of specific polymerase chain reaction product by utilizing the 5'----3' exonuclease activity of *Thermus aquaticus* DNA polymerase." *Proc. Natl. Acad. Sci. USA*, 88, 7276-7280.
52. Tyagi, S., and Kramer, F. R. (1996). "Molecular beacons: probes that fluoresce upon hybridization." *Nature Biotechnology*, 14, 303-308.
53. Tyagi, S., Bratu, D. P., and Kramer, F. R. (1998). "Multicolor molecular beacons for allele discrimination." *Nature Biotechnology*, 16, 49-53.
54. Bray, M. S., Boerwinkle, E., and Doris, P. A. (2001). "High-throughput multiplex SNP genotyping with MALDI-TOF mass spectrometry: practice, problems and promise." *Hum. Mutat.*, 17, 296-304.
55. Germer, S., and Higuchi, R. (1999). "Single-Tube genotyping without oligonucleotide probes." *Genome Res.*, 9, 72-78.

56. Hosfield, D. J., Frank, G., Weng, Y., Tainer, J. A., and Shen, B. (1998). "Newly discovered archaebacterial flap endonucleases show a structure-specific mechanism for DNA substrate binding and catalysis resembling human flap endonuclease-1." *J. Biol. Chem.*, 273, 27154-27161.
57. Kaiser, M. W., Lyamicheva, N., Ma, W., Miller, C., Neri, B., Fors, L., and Lyamichev, V. I. (1999). "A comparison of eubacterial and archael structure-specific 5'-exonucleases." *J. Biol. Chem.*, 30, 21387-21394.
58. Lyamichev, V., Mast, A. L., Hall, J. G., Prudent, J. R., Kaiser, M. W., Takova, T., Kwiatkowski, R. W., Sabder, T. J., de Arruda, M., Arco, D. A. et al. (1999). "Polymorphism identification and quantitative detection of genomic DNA by invasive cleavage of oligonucleotide probes." *Nature Biotechnology*, 17, 292-296.
59. <http://www.twt.com>
60. Mein, C. A., Barratt, B. J., Dunn, M. G., Siegmund, T., Smith, A. N., Esposito, L., Nutland, S., Stevens, H. E., Wilson, A. J., Phillips, M. S. et al. (2000). "Evaluation of single nucleotide polymorphism typing with invader on PCR amplicons and its automation." *Genome Res.*, 10, 330-343.
61. Ohnishi, Y., Tanaka, T., Ozaki, K., Yamada, R., Suzuki, H., and Nakamura, Y. (2001). "A high-throughput SNP typing system for genome-wide association studies." *J. Hum. Genet.*, 46, 471-477.
62. Karas, M., and Hillenkamp, F. (1988). "Laser desorption ionization of proteins with molecular masses exceeding 10000 daltons." *Anal. Chem.*, 60, 2299-2303.
63. Schlag, E. W., Grotemeyer, J., and Levine, R. D. (1992). "Do large molecules ionize?" *Chem. Phys. Lett.*, 190, 521-527.
64. Karas, M., Gluckmann, M., and Schafer, J. (2000). "Ionization in matrix-assisted laser desorption/ionization: singly charged molecular ions are the lucky survivors." *J. Mass Spectrom.*, 35, 1-12.
65. Hillenkamp, F., Karas, M., Beavis, R. C., and Chait, B. T. (1991). "Matrix-assisted laser desorption/ionization mass spectrometry of biopolymers." *Anal. Chem.*, 63, 1193A-1203A.

66. Yates, J. R., 3rd. (1998). "Mass spectrometry and the age of the proteome." *J. Mass Spectrom.*, 33, 1-19.
67. Monforte, J. A., and Becker, C. H. (1997). "High-throughput DNA analysis by time-of-flight mass spectrometry." *Nature Medicine*, 3, 360-362.
68. Colby, S. M., King, T.B., and Reilly, J.P. (1994). "Improving the resolution of matrix-assisted laser desorption/ionization time-of-flight mass spectrometry by exploiting the correlation between ion position and velocity." *Rapid Commun. Mass Spectrom.*, 8, 865-868.
69. Brown, R. S., and Lennon, J. J. (1995). "Mass resolution improvement by incorporation of pulsed ion extraction in a matrix-assisted laser desorption/ionization linear time-of-flight mass spectrometer." *Anal. Chem.*, 67, 1998-2003.
70. Berkenkamp, S., Kirpekar, F., and Hillenkamp, F. (1998). "Infrared MALDI Mass Spectrometry of Large Nucleic Acids." *Science*, 281, 260-262.
71. Griffin, T. J., and Smith, L. M. (2000). "Single-nucleotide polymorphism analysis by MALDI-TOF mass spectrometry." *Trends Biotechnol.*, 18, 77-84.
72. Smith, L. M. (1993). "The future of DNA sequencing." *Science*, 262, 530-532.
73. Kirpekar, F., Nordhoff, E., Larsen, L. K., Kristiansen, K., Roepstorff, P., and Hillenkamp, F. (1998). "DNA sequence analysis by MALDI mass spectrometry." *Nucleic Acids Res.*, 26, 2554-2559.
74. Fu, D. J., Tang, K., Braun, A., Reuter, D., Darnhofer-Demar, B., Little, D. P., O'Donnell, M. J., Cantor, C. R., and Koster, H. (1998). "Sequencing exons 5 to 8 of the p53 gene by MALDI-TOF mass spectrometry." *Nature Biotechnology*, 16, 381-384.
75. Nordhoff, E., Ingendoh, A., Cramer, R., Overberg, A., Stahl, B., Karas, M., Hillenkamp, F., and Crain, P. F. (1992). "Matrix-assisted laser desorption/ionization mass spectrometry of nucleic acids with wavelengths in the ultraviolet and infrared." *Rapid Commun. Mass Spectrom.*, 6, 771-776.
76. Mouradian, S., Rank, D. R., and Smith, L. M. (1996). "Analyzing sequencing reactions from bacteriophage M13 by matrix-assisted laser desorption/ionization mass spectrometry." *Rapid Commun. Mass Spectrom.*, 10, 1475-1478.

77. Taranenko, N. I., Allman, S. L., Golovlev, V. V., Taranenko, N. V., Isola, N. R., and Chen, C. H. (1998). "Sequencing DNA using mass spectrometry for ladder detection." *Nucleic Acids Res.*, 26, 2488-2490.
78. Gut, I. G., and Beck, S. (1995). "DNA and Matrix-assisted Laser Desorption Ionization Mass Spectrometry." *Molecular Biology: Current Innovations and Future Trends*, Wymondham, UK, 147-157.
79. Koster, H., Tang, K., Fu, D. J., Braun, A., van den Boom, D., Smith, C. L., Cotter, R. J., and Cantor, C. R. (1996). "A strategy for rapid and efficient DNA sequencing by mass spectrometry." *Nature Biotechnology*, 14, 1123-1128.
80. Ross, P., Davis, P., and Belgrader, P. (1998). "Analysis of DNA fragments from conventional and microfabricated PCR devices using delayed extraction MALDI-TOF mass spectrometry." *Anal. Chem.*, 70, 2067-2073.
81. Ross, P. L., and Belgrader, P. (1997). "Analysis of short tandem repeat polymorphisms in human DNA by matrix-assisted laser desorption/ionization mass spectrometry." *Anal. Chem.*, 69, 3966-3972.
82. Taranenko, N. I., Matteson, K. J., Chung, C. N., Zhu, Y. F., Chang, L. Y., Allman, S. L., Haff, L., Martin, S. A., and Chen, C. H. (1996). "Laser desorption mass spectrometry for point mutation detection." *Genet. Anal.*, 13, 87-94.
83. Haff, L. A., and Smirnov, I. P. (1997). "Multiplex genotyping of PCR products with MassTag-labeled primers." *Nucleic Acids Res.*, 25, 3749-3750.
84. Fei, Z., and Smith, L. M. (2000). "Analysis of single nucleotide polymorphisms by primer extension and matrix-assisted laser desorption/ionization time-of-flight mass spectrometry." *Rapid Commun. Mass Spectrom.*, 14, 950-959.
85. Haff, L. A., and Smirnov, I. P. (1997). "Single Nucleotide polymorphism identification assays using a thermostable DNA polymerase and delayed extraction MALDI-TOF mass spectrometry." *Genome Res.*, 7, 378-388.
86. Little, D. P., Braun, A., Darnhofer-Demar, B., and Koster, H. (1997). "Identification of apolipoprotein E polymorphisms using temperature cycled primer oligo base extension and mass spectrometry." *Eur. J. Clin. Chem. Clin. Biochem.*, 35, 545-548.



87. Little, D. P., Braun, A., O'Donnell, M. J., and Koster, H. (1997). "Mass spectrometry from miniaturized arrays for full comparative DNA analysis." *Nature Medicine*, 3, 1413-1416.
88. Ross, P., Hall, L., Smirnov, I., and Haff, L. (1998). "High level multiplex genotyping by MALDI-TOF mass spectrometry." *Nature Biotechnology*, 16, 1347-1351.
89. Li, J., Butler, J. M., Tan, Y., Lin, H., Royer, S., Ohler, L., Shaler, T. A., Hunter, J. M., Pollart, D. J., Monforte, J. A. et al. (1999). "Single nucleotide polymorphism determination using primer extension and time-of-flight mass spectrometry." *Electrophoresis*, 20, 1258-1265.
90. Nordhoff, E., Luebbert, C., Thiele, G., Heiser, V., and Lehrach, H. (2000). "Rapid determination of short DNA sequences by the use of MALDI-MS." *Nucleic Acids Res.*, 28, E86.
91. Tang, K., Fu, D.-J., Julien, D., Braun, A., Cantor, C. R., and Koester, H. (1999). "Chip-based genotyping by mass spectrometry." *Proc. Natl. Acad. Sci. USA*, 96, 10016-10020.
92. <http://www.sequenom.com>
93. <http://home.appliedbiosystems.com/>
94. Egholm, M., Buchardt, O., Christensen, L., Behrens, C., Freier, S. M., Driver, D. A., Berg, R. H., Kim, S. K., Norden, B., and Nielsen, P. E. (1993). "PNA hybridizes to complementary oligonucleotides obeying the Watson-Crick hydrogen-bonding rules." *Nature*, 365, 566-568.
95. Tomac, S., Sarkar, M., Ratilainen, T., Wittung, P., Nielson, P. E., Nordén, B., and Gräslund, A. (1996). "Ionic effects on the stability and conformation of peptide nucleic acid complexes." *J. Am. Chem. Soc.*, 118, 5544-5552.
96. Mollegaard, N. E., Buchardt, O., Egholm, M., and Nielsen, P. E. (1994). "Peptide nucleic acid.DNA strand displacement loops as artificial transcription promoters." *Proc. Natl. Acad. Sci. USA*, 91, 3892-3895.

97. Griffin, T. J., Tang, W., and Smith, L. M. (1997). "Genetic analysis by peptide nucleic acid affinity MALDI-TOF mass spectrometry." *Nature Biotechnology*, 15, 1368-1372.
98. Ross, P. L., Lee, K., and Belgrader, P. (1997). "Discrimination of single-nucleotide polymorphisms in human DNA using peptide nucleic acid probes detected by MALDI-TOF mass spectrometry." *Anal. Chem.*, 69, 4197-202.
99. Griffin, T. J., and Smith, L. M. (1998). "An approach to predicting the stabilities of peptide nucleic acid:DNA duplexes." *Anal. Biochem.*, 260, 56-63.
100. Giesen, U., Kleider, W., Berding, C., Geiger, A., Orum, H., and Nielsen, P. E. (1998). "A formula for thermal stability (T<sub>m</sub>) prediction of PNA/DNA duplexes." *Nucleic Acids Res.*, 26, 5004-5006.
101. Stoerker, J., Mayo, J. D., Tetzlaff, C. N., Sarracino, D. A., Schwoppe, I., and Richert, C. (2000). "Rapid genotyping by MALDI-monitored nuclease selection from probe libraries." *Nature Biotechnology*, 18, 1213-1216.
102. <http://www.qiagenomics.com>
103. Griffin, T. J., Hall, J. G., Prudent, J. R., and Smith, L. M. (1999). "Direct genetic analysis by matrix-assisted laser desorption/ionisation mass spectrometry." *Proc. Natl. Acad. Sci. USA*, 96, 6301-6306.
104. Griffin, T. J., and Smith, L. M. (2000). "Genetic identification by mass spectrometric analysis of single-nucleotide polymorphisms: ternary encoding of genotypes." *Anal. Chem.*, 72, 3298-3302.
105. Christian, N. P., Colby, S. M., Giver, L., Houston, C. T., Arnold, R. J., Ellington, A. D., and Reilly, J. P. (1995). "High resolution matrix-assisted laser desorption/ionization time-of-flight analysis of single-stranded DNA of 27 to 68 nucleotides in length." *Rapid Commun. Mass Spectrom.*, 9, 1061-1066.
106. Pieves, U., Zuercher, W., Schaer, M., and Moser, H. E. (1993). "Matrix-assisted laser desorption time-of-flight mass spectrometry : a powerful tool for the mass and sequence analysis of natural and modified oligonucleotides." *Nucleic Acids Res.*, 14, 3191-3196.

107. Nordhoff, E., Kirpekar, F., Karas, M., Cramer, R., Hahner, S., Hillenkamp, F., Kristiansen, K., Roepstorff, P., and Lezius, A. (1994). "Comparison of IR- and UV-matrix-assisted laser desorption/ionization mass spectrometry of oligodeoxynucleotides." *Nucleic Acids Res.*, 22, 2460-2465.
108. Kirpekar, F., Nordhoff, E., Kristiansen, K., Roepstorff, P., Hahner, S., and Hillenkamp, F. (1995). "7-Deaza purine bases offer a higher ion stability in the analysis of DNA by matrix-assisted laser desorption/ionization mass spectrometry." *Rapid Commun. Mass Spectrom.*, 9, 525-531.
109. Schneider, K., and Chait, B. T. (1995). "Increased stability of nucleic acids containing 7-deaza-guanosine and 7-deaza-adenosine may enable rapid DNA sequencing by matrix-assisted laser desorption mass spectrometry." *Nucleic Acids Res.*, 23, 1570-1575.
110. Nordhoff, E., Cramer, R., Karas, M., Hillenkamp, F., Kirpekar, F., Kristiansen, K., and Roepstorff, P. (1993). "Ion stability of nucleic acids in infrared matrix-assisted laser desorption/ionization mass spectrometry." *Nucleic Acids Res.*, 21, 3347-3357.
111. Gut, I. G., and Beck, S. (1995). "A procedure for selective DNA alkylation and detection by mass spectrometry." *Nucleic Acids Res.*, 23, 1367-1373.
112. Keough, T., Baker, T. R., Dobson, R. L., Lacey, M. P., Riley, T. A., Hasselfield, J. A., and Hesselberth, P. E. (1993). "Antisense DNA oligonucleotides. II: The use of matrix-assisted laser desorption/ionization mass spectrometry for the sequence verification of methylphosphonate oligodeoxyribonucleotides." *Rapid Commun. Mass Spectrom.*, 7, 195-200.
113. Bartlet-Jones, M., Jeffery, W. A., Hansen, H. F., and Pappin, D. J. (1994). "Peptide ladder sequencing by mass spectrometry using a novel, volatile degradation reagent." *Rapid Commun. Mass Spectrom.*, 8, 737-42.
114. Gut, I. G., Jeffery, W. A., Pappin, D. J. C., and Beck, S. (1997). "Analysis of DNA by 'Charge Tagging' and Matrix-assisted Laser Desorption /Ionization Mass Spectrometry." *Rapid Commun. Mass Spectrom.*, 11, 43-50.

115. Berlin, K., and Gut, I. G. (1999). "Analysis of Negatively 'Charge Tagged' DNA by Matrix-assisted Laser Desorption/Ionization Mass Spectrometry." *Rapid Commun. Mass Spectrom.*, 13, 1739-1743.
116. Sambrook, J., Fritsch, E., and Maniatis, T. (1989). "Molecular Cloning: a laboratory manual." *Cold Spring Harbor Press, New York, USA*.
117. Seela, F., and Zulauf, M. (1999). "Oligonucleotides Containing 7-Deazaadenines: The Influence of the 7-Substituent Chain Length and Change on the Duplex Stability." *Helv. Chim. Acta*, 82, 1878-1898.
118. Ramzaeva, N., Mittelbach, C., and Seela, F. (1997). "7-Deazaguanine DNA: Oligonucleotides with Hydrophobic or Cationic Side Chains." *Helv. Chim. Acta*, 80, 1809-1822.
119. Ramzaeva, N., Mittelbach, C., and Seela, F. (1999). "7-Deaza-2'-Deoxyguanosines Functionalized With 7-( $\omega$ -Aminoalkyl-1-ynyl) Residues." *Nucleosides & Nucleotides*, 18, 1439-1440.
120. Robbins, M., and Barr, P. (1983). "Nucleic Acid Related Compounds. Efficient Conversion of 5-Iodo to 5-Alkenyl and Derived 5-Substituted Uracil Bases and Nucleosides." *J. Org. Chem.*, 1854-1862.
121. Pusch, W., Kraeuter, K. O., Froehlich, T., Stalgies, Y., and Kostrzewa, M. (2001). "Genotools SNP manager: a new software for automated high-throughput MALDI-TOF mass spectrometry SNP genotyping." *Biotechniques*, 30, 210-215.
122. <http://www.cng.fr>
123. Rao, H. G., Rosenfeld, A., and Wetmur, J. G. (1998). "Methanococcus jannaschii flap endonuclease: expression, purification, and substrate requirements." *J. Bacteriol.*, 180, 5406-5412.
124. Sauer, S., Lechner, D., Berlin, K., Lehrach, H., Escary, J. L., Fox, N., and Gut, I. G. (2000). "A novel procedure for efficient genotyping of single nucleotide polymorphisms." *Nucleic Acids Res.*, 28, E13.

125. Sauer, S., Lechner, D., Berlin, K., Plançon, C., Heuermann, A., Lehrach, H., and Gut, I. G. (2000). "Full flexibility genotyping of single nucleotide polymorphisms by the GOOD assay." *Nucleic Acids Res.*, 28, E100.
126. Sauer, S., and Gut, I. G. "Enhancements of the GOOD assay for genotyping single nucleotide polymorphisms by MALDI-MS.", submitted
127. Collinge, J., Sidle, K. C. L., Meads, J., Ironside, J., and Hill, A. F. (1996). "Molecular analysis of prion strain variation and the aetiology of "new variant" CJD." *Nature*, 383, 685-690.
128. <http://genecanvas.idf.inserm.fr>
129. Soubrier, F., Nadaud, S., and Williams, T. A. (1994). "Angiotensin I converting enzyme gene: regulation, polymorphism and implications in cardiovascular diseases." *Eur. Heart J.*, 15 Suppl D, 24-29.
130. Newman, P. J. (1997). "The biology of PECAM-1." *J. Clin. Invest.*, 100 (11 Suppl), S 25-29.
131. Adachi, T., and Alam, R. (1998). "The mechanism of IL-5 signal transduction." *Am. J. Physiol.*, 275, 623-633.
132. Fossati, G., Mazzucchelli, I., Gritti, D., Ricevuti, G., Edwards, S. W., Moulding, D. A., and Rossi, M. L. (1998). "In vitro effects of GM-CSF on mature peripheral blood neutrophils." *Int. J. Mol. Med.*, 943-951.
133. Reynolds, M. A., Hogrefe, R. I., Jaeger, J. A., Schwartz, D. A., Riley, T. A., Marvin, W. B., Daily, W. J., Vaghefi, M. M., Beck, T. A., Knowles, S. K. et al. (1996). "Synthesis and thermodynamics of oligonucleotides containing chirally pure R(P) methylphosphonate linkages." *Nucleic Acids Res.*, 24, 4584-4591.
134. Ellis, L. A., Taylor, C. F., and Taylor, G. R. (2000). "A comparison of fluorescent SSCP and denaturing HPLC for high throughput mutation scanning." *Hum. Mutat.*, 15, 556-564.
135. Sauer, S. Gelfand, D. H., Boussicault, F., Bauer, K., Reichert, F., and Gut, I. G. "Facile method for automated genotyping of single nucleotide polymorphisms.", submitted

136. Agrawal, S. (1992). "Antisense oligonucleotides as antiviral agents." *Trends Biotechnol.*, 10, 152-158.
137. Timmermann, B., Mo, R., Luft, F. C., Gerds, E., Busjahn, A., Omvik, P., Li, G. H., Schuster, H., Wienker, T. F., Hoehe, M. R. et al. (1998). "Beta-2 adrenoceptor genetic variation is associated with genetic predisposition to essential hypertension: The Bergen Blood Pressure Study." *Kidney Int.*, 53, 1455-1460.
138. Busjahn, A., Li, G. H., Faulhaber, H. D., Rosenthal, M., Becker, A., Jeschke, E., Schuster, H., Timmermann, B., Hoehe, M. R., and Luft, F. C. (2000). "beta-2 adrenergic receptor gene variations, blood pressure, and heart size in normal twins." *Hypertension*, 35, 555-560.
139. Jurinke, C., van den Boom, D., Jacob, A., Tang, K., Woehrl, R., and Koester, H. (1996). "Analysis of ligase chain reaction products via matrix-assisted laser desorption/ionisation mass spectrometry." *Anal. Biochem.*, 237, 174-181.
140. Eis, P. S., Olson, M. C., Takova, T., Curtis, M. L., Olson, S. M., Vener, T. I., Ip, H. S., Vedvik, K. L., Bartholomay, C. T., Allawi, H. T., Ma, W. P., Hall, J. G., Morin, M. D., Rushmore, T. H., Lyamichev, V. I., and Kwiatkowski, R. W. (2001). "An invasive cleavage assay for direct quantitation of specific RNAs." *Nature Biotechnology*, 19, 673-676.
141. Lyamichev, V. I., Kaiser, M. W., Lyamicheva, N. E., Vologodskii, A. V., Hall, J. G., Ma, W. P., Allawi, H. T., and Neri, B. P. (2000). "Experimental and theoretical analysis of the invasive signal amplification reaction." *Biochemistry*, 39, 9523-9532.
142. Gut, I. G., Lechner, D., and Sauer, S. (2000). "Sample generation for genotyping by mass spectrometry", submitted to the European Patent Office, The Hague, Netherlands
143. <http://www.millipore.com>
144. <http://www.qiagen.com>
145. Mullis, K., Faloona, F., Scharf, S., Saiki, R., Horn, G., and Erlich, H. (1986). "Specific enzymatic amplification of DNA in vitro: the polymerase chain reaction." *Cold Spring Harb. Symp. Quant. Biol.*, 51 Pt. 1, 263-73.

146. Mullis, K. B., and Faloona, F. A. (1987). "Specific synthesis of DNA in vitro via a polymerase-catalyzed chain reaction." *Methods in Enzymology*, 155, 335-350.
147. Schatz, P. (2000). "Standardisierung einer SNP Genotypisierung mittels Massenspektrometrie." Diploma Thesis at the Technical University Berlin
148. Ross, P., Hall, L., and Haff, L. A. (2000). "Quantitative approach to single-nucleotide polymorphism analysis using MALDI-TOF mass spectrometry." *Biotechniques*, 29, 620-626, 628-629.
149. Douglas, J. A., Boehnke, M., Gillanders, E., Trent, J. M., and Gruber, S. B. (2001). "Experimentally-derived haplotypes substantially increase the efficiency of linkage disequilibrium studies." *Nature Genetics*, 28, 361-364.
150. Brandt, O. (2000). "Haplotypisierung mittels Massenspektrometrie." Diploma Thesis at the Technical University Berlin
151. Ruano, G., and Kidd, K. K. (1989). "Direct haplotyping of chromosomal segments from multiple heterozygotes via allele-specific PCR amplification." *Nucleic Acids Res.*, 17, 8392.
152. Pretsch, E., Clerc, T., Seibl, J., and Simon, W. (1981). "Tabellen zur Strukturaufklärung organischer Verbindungen mit spektroskopischen Methoden." *Springer-Verlag Berlin Heidelberg New York*.
153. Sauer, S. Lechner, D., and Gut I.G. (2001). "The GOOD assay." *Mass Spectrometry and Genomic Analysis, Kluwer Academic Publishers, Netherlands*.
154. Luo, J., Bergstrom, D. E., and Barany, F. (1996). "Improving the fidelity of *Thermus thermophilus* DNA ligase." *Nucleic Acids Res.*, 24, 3071-3078.
155. Birkenmeyer, L., and Armstrong, A. S. (1992). "Preliminary evaluation of the ligase chain reaction for specific detection of *Neisseria gonorrhoeae*." *J. Clin. Microbiol.*, 30, 3089-3094.
156. Wiedemann, M., Czaika, J., Barany, F., and Batt, C. A. (1992). "Discrimination of *Listeria monocytogenes* from other *Listeria* species by ligase chain reaction." *Appl. Environ. Microbiol.*, 58, 3443-3447.

157. Nilsson, M., Malmgren, H., Samoitaki, M., Kwiatkowski, M., Chowdhary, B. P., and Landegren, U. (1994). "Padlock Probes : Circulizing oligonucleotides for localized DNA detection." *Science*, 265, 2085-2088.
158. Christian, N.P., personal communication
159. Gut, I.G., personal communication
160. Buetow, K. H., Edmonson, M., MacDonald, R., Clifford, R., Yip, P., Kelley, J., Little, D. P., Strausberg, R., Koester, H., Cantor, C. R. et al. (2001). "High-throughput development and characterization of a genomewide collection of gene-based single nucleotide polymorphism markers by chip-based matrix-assisted laser desorption/ionization time-of-flight mass spectrometry." *Proc. Natl. Acad. Sci. USA*, 98, 581-584

### Abbreviations

$\alpha$ -S-dATP = 2'-Deoxyadenosine-5'-O-(1-Thiotriphosphate)

$\alpha$ -S-dCTP = 2'-Deoxycytidine-5'-O-(1-Thiotriphosphate)

$\alpha$ -S-dGTP = 2'-Deoxyguanosine-5'-O-(1-Thiotriphosphate)

$\alpha$ -S-dNTP = 2'-Deoxynucleoside-5'-O-(1-Thiotriphosphate)

$\alpha$ -S-dTTP = 2'-Deoxythymidine-5'-O-(1-Thiotriphosphate)

$\alpha$ -S-ddATP = 2',3' Dideoxyadenosine-5'-O-(1-Thiophosphate)

$\alpha$ -S-ddCTP = 2',3' Dideoxycytidine-5'-O-(1-Thiophosphate)

$\alpha$ -S-ddGTP = 2',3' Dideoxyguanosine-5'-O-(1-Thiophosphate)

$\alpha$ -S-ddNTP = 2',3' Dideoxynucleoside-5'-O-(1-Thiophosphate)

$\alpha$ -S-ddTTP = 2',3' Dideoxythymidine-5'-O-(1-Thiophosphate)

A = Adenine

A<sub>600</sub> = Absorption at 600 nm



## Appendix

A<sup>NH2</sup> = Propargylaminomodified 7-deaza-2'-deoxyadenosine

ACE = Angiotensin converting enzyme

AEBSF = 4-(2-Aminoethyl)-benzenesulfonyl fluoride, hydrochloride

BRCA = Breast-cancer associated

BSE = Bovine spongiform encephalopathy

C = Cytosine

CAV = Caveolin

CCD = Charge-coupled device

cDNA = complementary DNA

CJD = Creutzfeldt-Jakob disease

CNG = Centre National de Génotypage

CNME =  $\alpha$ -Cyano-4-hydroxy-cinnamic acid methyl ester

C<sup>NH2</sup> = 5-propargylaminomodified 2'-deoxycytidine

CT = Charge tag

dATP = 2'-Deoxyadenosine-5'-Triphosphate

dCTP = 2'-Deoxycytidine-5'-Triphosphate

dGTP = 2'-Deoxyguanosine-5'-Triphosphate

dNTP = 2'-Deoxynucleoside-5'-Triphosphate

dTTP = 2'-Deoxythymidine-5'-Triphosphate

ddATP = 2',3'-Dideoxyadenosine-5'-Triphosphate

ddCTP = 2',3'-Dideoxycytidine-5'-Triphosphate

ddGTP = 2',3'-Dideoxyguanosine-5'-Triphosphate

ddNTP = 2',3'-Dideoxynucleosid-5'-Triphosphate

ddTTP = 2',3'-Dideoxythymidine-5'-Triphosphate

Da = Dalton

## Appendix

DNA = Deoxyribonucleic acid

DTT = Dithiothreitol

E.coli = Escherichia coli

EDTA = Ethylenediaminetetraacetic acid

FEN = Flap endonuclease

FPLC = Fast performance liquid chromatography

G = Guanine

GM-SCF = Granulocyte-macrophage colony stimulating factor

G<sup>NH2</sup> = Propargylaminommodified 7-deaza-2'-deoxyguanosine

g = Gram

HNPPC = Hereditary nonpolyposis colectral cancer

HPA = Hydroxy picolinic acid

HPLC = High performance liquid chromatography

HPSF = High purity salt free

h = Hour

LB = Luria broth

LCR = Ligase chain reaction

LIMS = Laboratory information management system

l = Liter

M = Molar

MALDI = Matrix-assisted laser desorption/ionisation

MALDI-MS = Matrix-assisted laser desorption/ionisation mass spectrometry

ml = Milliliter

mM = Minimolar

MODY = Maturity-onset diabetes of the young

## Appendix

MOPS = 3-[N-Morpholino]propanesulfonic acid

mp = Methylphosphonate

m/z = Mass to charge ratio

NAD = ( $\beta$ -) Nicotinamide adenine dinucleotide

OD = Optical density

PAGE = Polyacrylamide gel electrophoresis

PCR = Polymerase chain reaction

PECAM = Platelet/endothelial cell adhesion molecule

pmol = Picomol

PT = Phosphorothioate

rpm = Revolution per minute

SNP = Single nucleotide polymorphism

T = Thymine

TBE = Tris-Borate-EDTA

TE = Tris-EDTA

T<sup>NH2</sup> = Amino modifier C6 dT

TOF = Time-of-flight

Tris = Tris(hydroxymethyl)aminomethane

U = Units

U = Uridine

U<sup>NH2</sup> = 5-propargylaminommodified 2'-deoxyuridine

UV = Ultraviolet

$\mu$ l = Microliter

$\mu$ M = Micromolar

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S. Sauer and I.G. Gut: “Enhancements of the GOOD assay for genotyping single nucleotide polymorphisms by MALDI-MS”, submitted

S. Sauer, D. Lechner, K. Berlin, C. Plançon, A. Heuermann, H. Lehrach and I. G. Gut: “Full flexibility genotyping of single nucleotide polymorphisms by the GOOD assay“, *Nucleic Acids Res.* 28, e100, 2000

S. Sauer, D. Lechner, K. Berlin, H. Lehrach, J.-L. Escary, N. Fox and I.G. Gut: “A novel procedure for efficient genotyping of single nucleotide polymorphisms“, *Nucleic Acids Res.* 28, e13, 2000

Patent:

I.G. Gut, D. Lechner and S. Sauer: “Sample Generation for genotyping by mass spectrometry”, submitted to the European Patent Office

Book chapter:

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