

Development of
Genome-Probing Microarrays for
Monitoring Lactic Acid Bacteria during Kimchi
Fermentation

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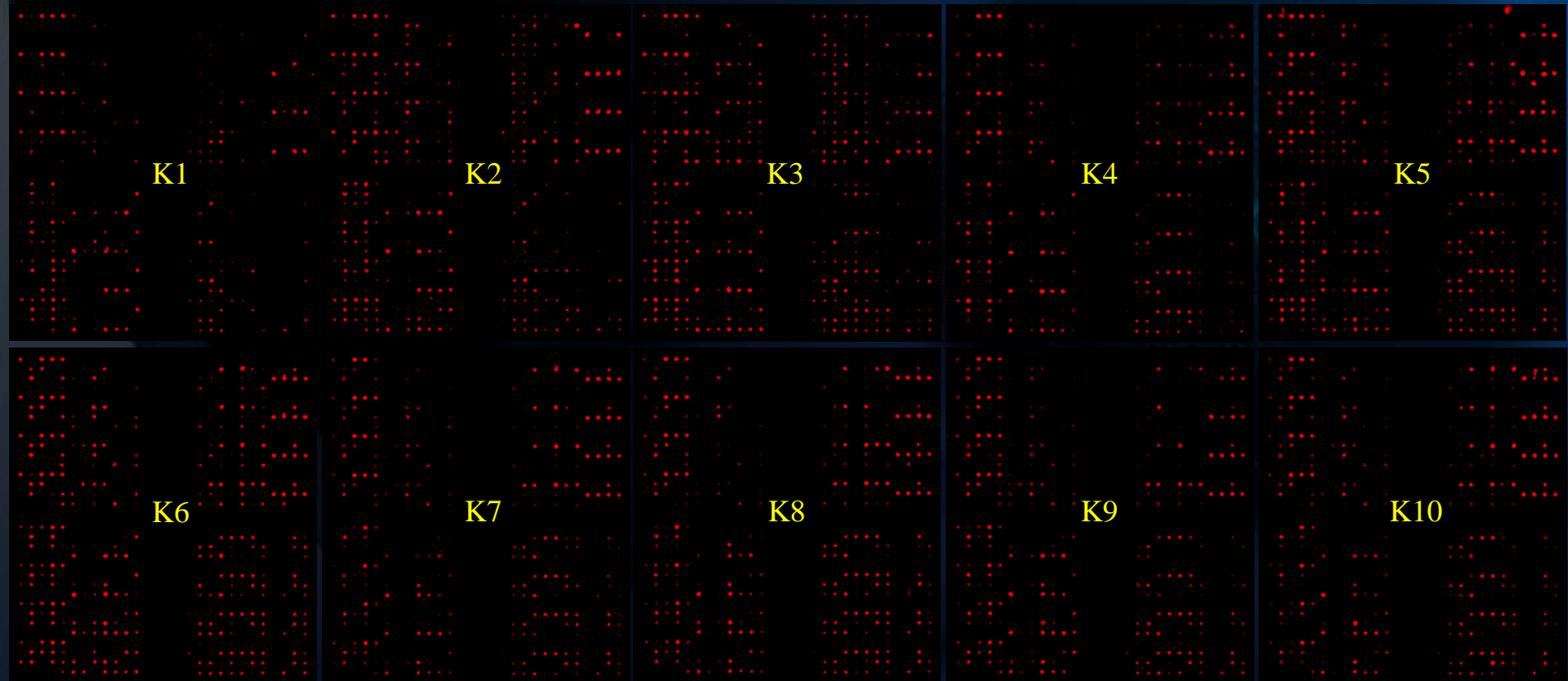
Various types of microbial diagnostic microarrays

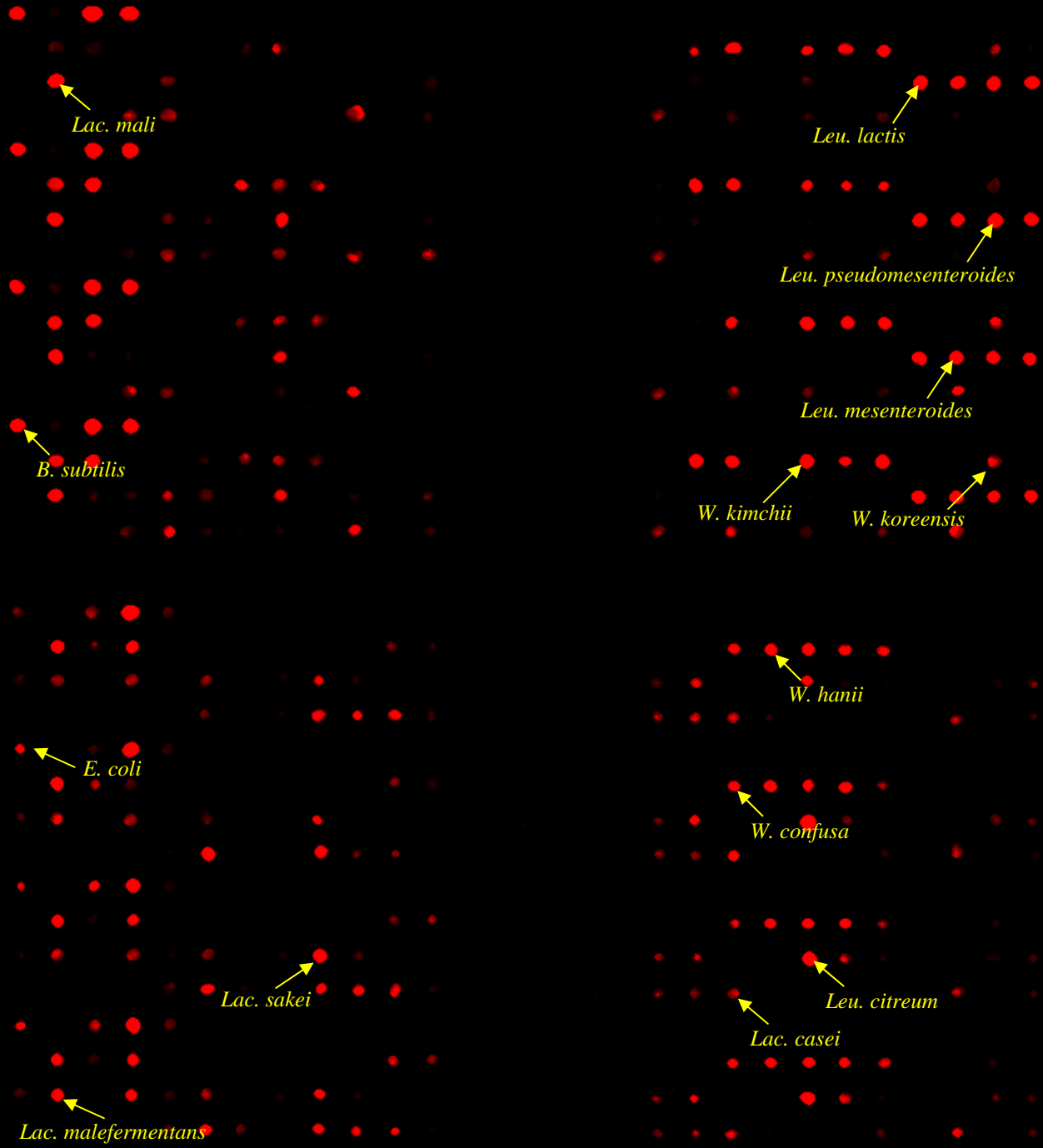
	Probe Size	Sensitivity	Fabrication method	Advantage	Disadvantage
Oligomer chip	20-70bp	25ng	<i>in silico</i> design & Oligomer synthesizer	Detect not-isolated strains & low cost for probe design	Low sensitivity & specificity
cDNA microarray	0.2 – 0.5 Kbp	2.5ng	<i>in silico</i> primer design & PCR	Detect not-isolated strains & high sensitivity	Extremely low specificity
Genome probing	3-5 Mbp	0.25ng	Culture & Genome extraction	High sensitivity & specificity	Low throughput obtaining of genome & only detect isolates

Target Microorganisms : Lactic Acid Bacteria

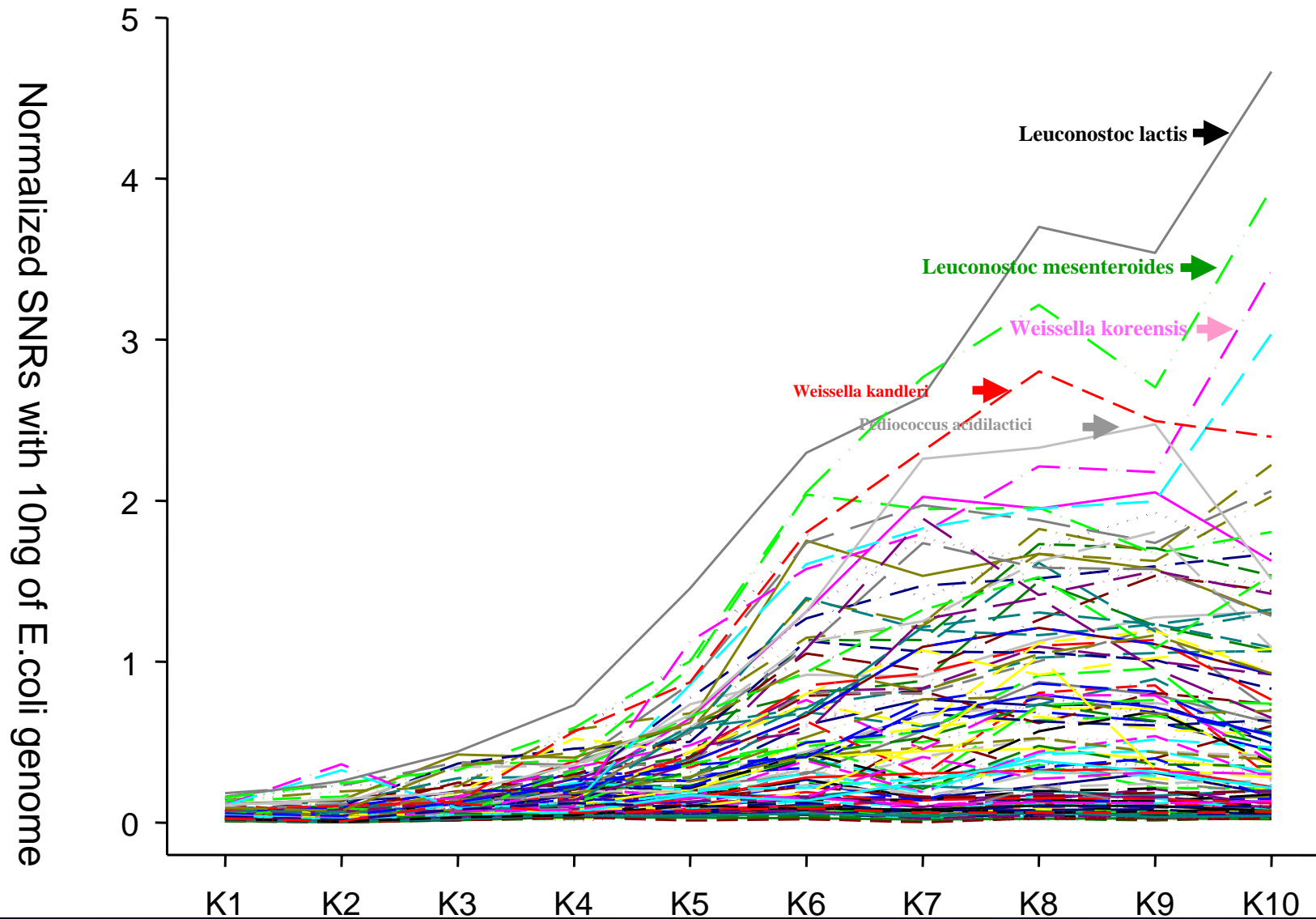
Bif. angulatum	Bif. bifidum	Bif. coryneforme	Bif. minimum	Bif. saeculare	Car. divergens	Car. piscicola	Ent. cecorum	Ent. durans	Ent. faecium	Ent. flavesceus	Ent. haemoferroxidus	Ent. hirae	Ent. malodoratus	Ent. moraviensis	Ent. mundtii	Ent. phoeniculicola	Ent. saccharolyticus	Ent. solitarius	Ent. villorum
Lac. acidophilus	Lac. agilis	Lac. alimentarius	Lac. amylophilus	Lac. amylovorus	Lac. animalis	Lac. brevis	Lac. casei	Lac. coryniformis sub. coryniformis	Lac. coryniformis sub. torquens	Lac. crispatus	Lac. curvatus	Lac. cypricaei	Lac. delbrueckii sub. bulgaricus	Lac. delbrueckii sub. delbrueckii	Lac. delbrueckii sub. lactis	Lac. diolivorans	Lac. equi	Lac. farciminis	Lac. fermentum
Lac. fructivorans	Lac. fuchuensis	Lac. graminis	Lac. helveticus	Lac. ingluviei	Lac. jensenii	Lac. johnsonii	Lac. kefirii	Lac. kimchi	Lac. malefermentans	Lac. mali	Lac. maltaromicus	Lac. murinus	Lac. oris	Lac. parabucchneri	Lac. paracasei sub. paracasei	Lac. paracasei sub. tolerans	Lac. pentosus	Lac. plantarum	Lac. reuteri
Lac. rhamnosus	Lac. ruminis	Lac. sakei sub. carnosus	Lac. sakei sub. sakei	Lac. salivarius sub. salicinius	Lac. sanfranciscensis	Lac. sharpeae	Lac. suebicus	Lac. vaccinostercus	Lac. vaginalis	Lac. vitulinus	Lac. zaeae	Lcc. garvieae	Lcc. lactis sub. hordniae	Lcc. lactis sub. lactis	Lcc. piscium	Lcc. plantarum	Lcc. raffinolactis	Leu. argentinum	Leu. carnosum
Leu. citreum	Leu. citreum	Leu. fallax	Leu. fructosum	Leu. gelidum	Leu. inhae	Leu. lactis	Leu. mesenteroides sub. cremoris	Leu. mesenteroides sub. dextranicum	Leu. mesenteroides sub. mesenteroides	Leu. pseudomesenteroides	Oenococcus oeni	Pedio. acidilactici	Pedio. claussenii	Pedio. damnosus	Pedio. dextrinicus	Pedio. inopinatus	Pedio. parvulus	Pedio. pentosaceus	Pedio. urinaequi
Strepto. alactolyticus	Strepto. anginosus	Strepto. australis	Strepto. criceti	Strepto. downei	Strepto. entericus	Strepto. equi sub. zooepidemicus	Strepto. ferus	Strepto. gordonii	Strepto. hyointestinalis	Strepto. iniae	Strepto. mitis	Strepto. mutans	Strepto. pyogenes	Strepto. rattii	Strepto. sanguinis	Strepto. sobrinus	Strepto. suis	Strepto. thermophilus	Strepto. vestibularis
Weiss. cibaria	Weiss. confusa	Weiss. halotolerans	Weiss. hani	Weiss. hellenica	Weiss. kandleri	Weiss. kimchii	Weiss. minor	Weiss. paramesenteroides	Weiss. soli	Weiss. thailandensis	Weiss. viridescens	MR2	MR5	MR6	KM1	KM4	KM6	Negative Control	Negative Control

Monitoring Population Dynamics of Lactic Acid Bacteria during Fermentation of Kimchi using Genome-Probing Microarrays





Population Dynamics of Lactic Acid Bacteria during Fermentation of Kimchi



Development and Evaluation of Genome-Probing Microarrays for Monitoring Lactic Acid Bacteria†

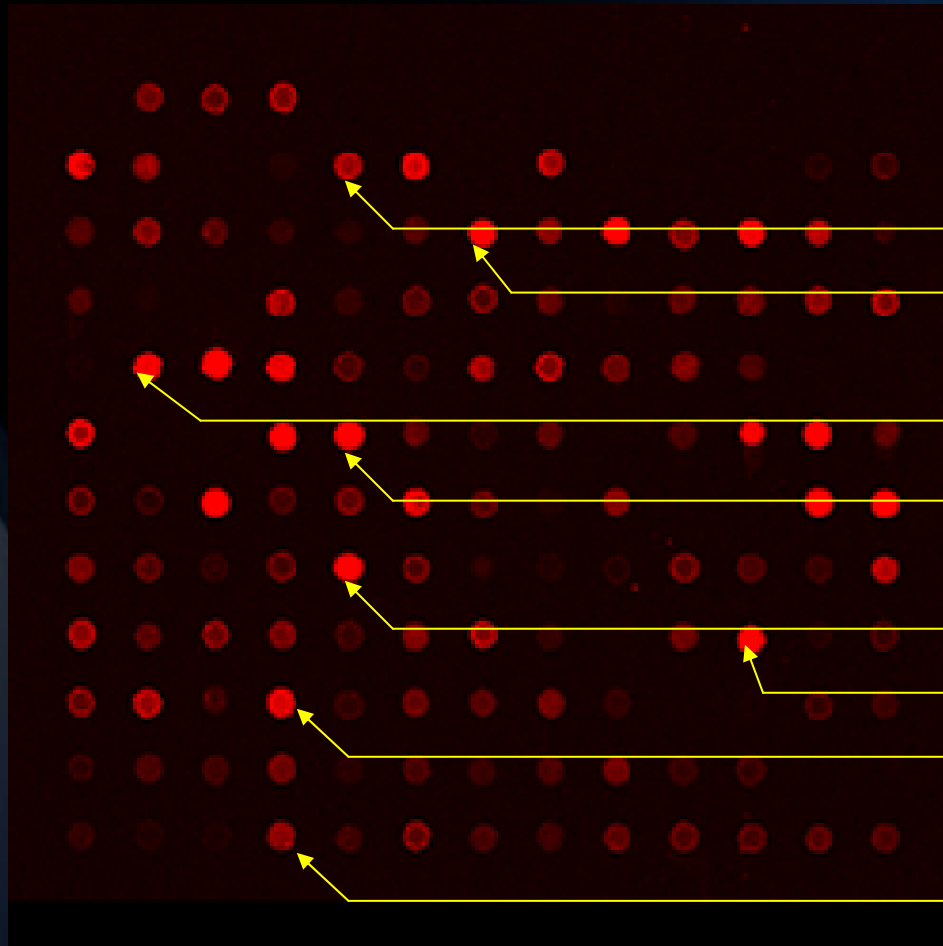
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The genome-probing microarray (GPM) was developed for quantitative, high-throughput monitoring of community dynamics in lactic acid bacteria (LAB) fermentation through the deposit of 149 microbial genomes as probes on a glass slide. Compared to oligonucleotide microarrays, the specificity of GPM was remarkably increased to a species-specific level. GPM possesses about 10- to 100-fold higher sensitivity (2.5 ng of genomic DNA) than the currently used 50-mer oligonucleotide microarrays. Since signal variation between the different genomes was very low compared to that of cDNA or oligonucleotide-based microarrays, the capacity of global quantification of microbial genomes could also be observed in GPM hybridization. In order to assess the applicability of GPMs, LAB community dynamics were monitored during the fermentation of kimchi, a traditional Korean food. In this work, approximately 100 diverse LAB species could be quantitatively analyzed as actively involved in kimchi fermentation.

Microarray profiling of starter cultured kimchii



Weissella cibaria

Weissella confusa

Leuconostoc lactis

Leuconostoc mesenteroides
subsp. *Mesenteroides*

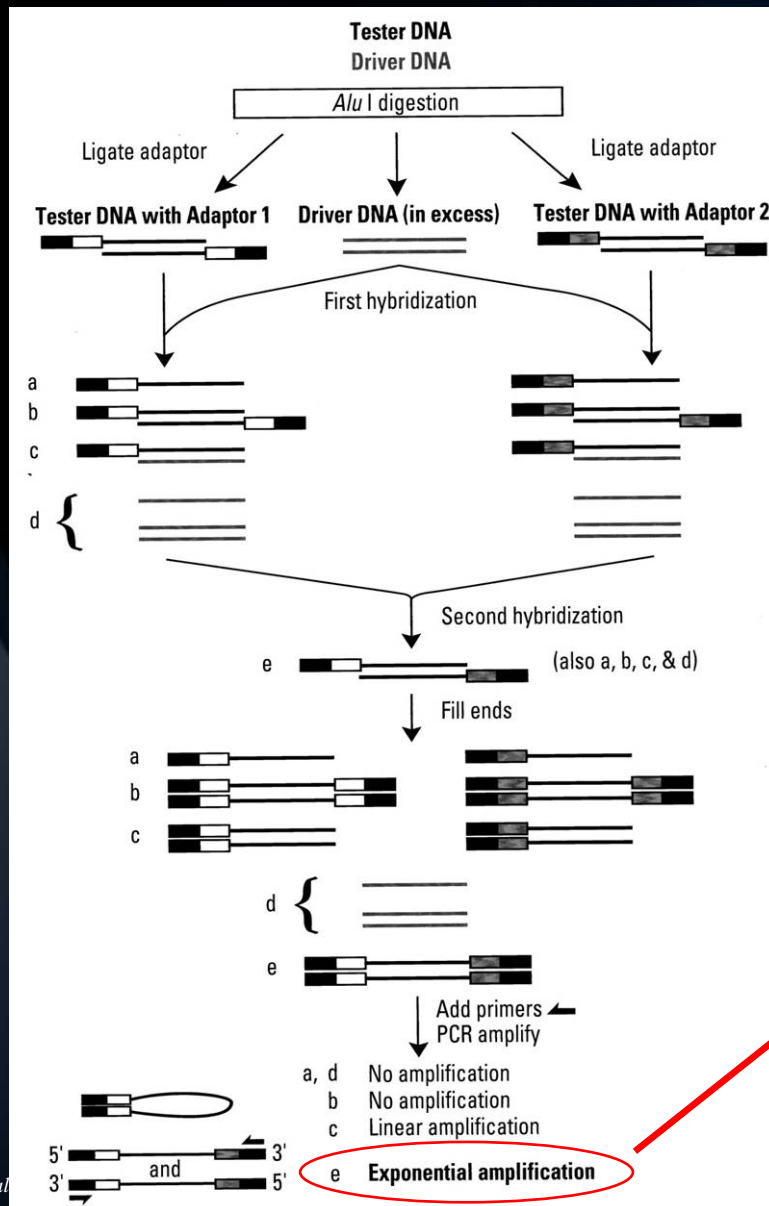
Lactobacillus sakei subsp. *sakei*

Lactobacillus malefermentans

Lactobacillus delbrueckii subsp. *bulgari*

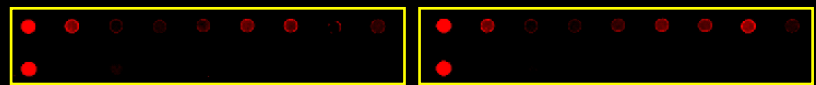
Carnobacterium divergens

Subtracted Genome Microarrays (SGMs)

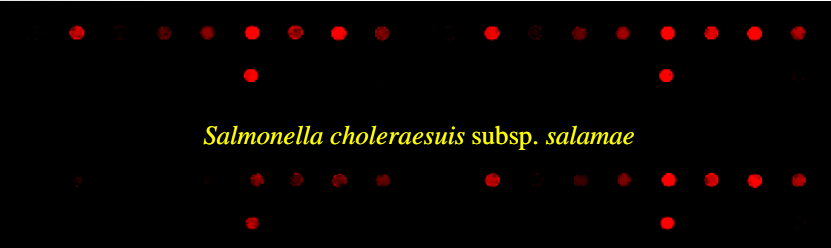


Fabrication of microarrays
with strain (or subspecies)
- level resolution

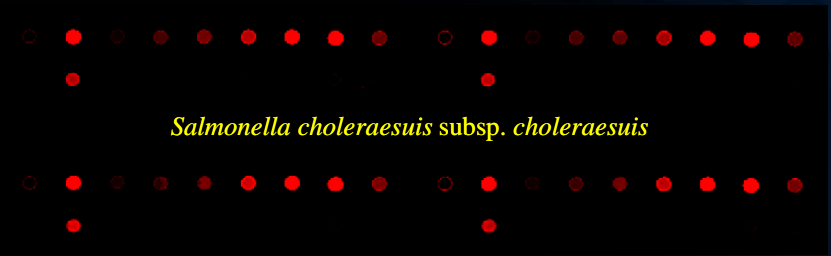
Printing the strain-specific genomes
using PCR-based **suppression**
subtractive hybridization



Salmonella choleraesuis subsp. *indica*



Salmonella choleraesuis subsp. *salamae*



Salmonella choleraesuis subsp. *choleraesuis*



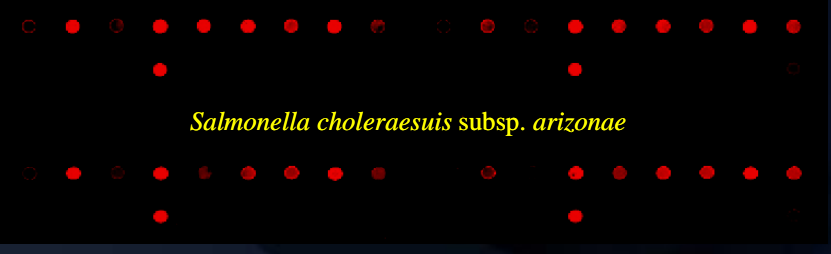
Salmonella typhimurium



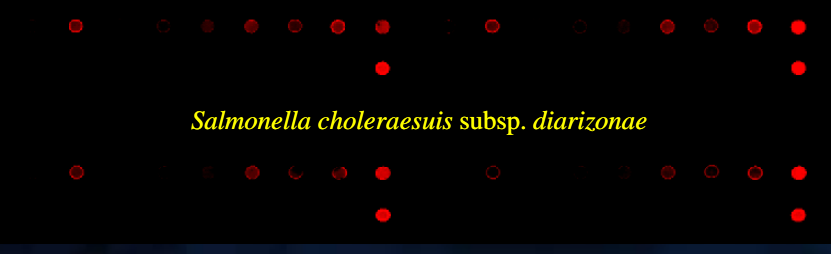
Salmonella bongori



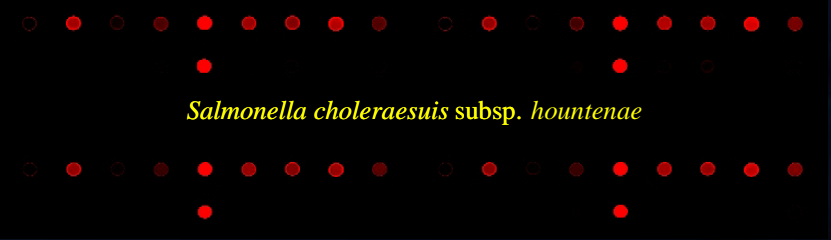
Salmonella enteritidis



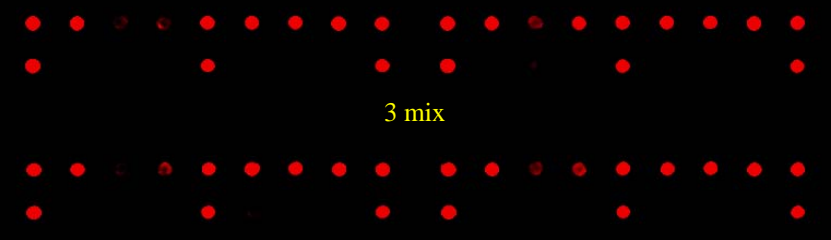
Salmonella choleraesuis subsp. *arizonae*



Salmonella choleraesuis subsp. *diarizonae*



Salmonella choleraesuis subsp. *hountenae*



3 mix

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Generation of subspecies level-specific microbial diagnostic microarrays using genes amplified from subtractive suppression hybridization as microarray probes

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