



CONFIDENTIAL

**Analysis of
Sigmund Gjernes Voss
yeast sample
for
Lars Marius Garshol
RÆLINGEN
NORWAY
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Interpretation of Results

The fingerprinting technique gives rise to DNA fragments of various lengths. The distribution of fragment lengths will vary from strain to strain. To visualise the DNA fragments the mixture is loaded onto a 1.5% agarose gel and electrophoretically separated. The rate of migration through the gel depends on size. The smaller the piece of DNA the further it migrates. Following staining the DNA can be visualised and photographed under UV illumination. These Photograph(s) are included in the report.

Normally there will be between 5-20 bands per track, depending on the strain. The differences between strains are shown as the presence or absence of bands of specific sizes. Closely related strains will have a number of bands in common, while unrelated strains will have totally different banding patterns.

The presence or absence of bands reflect a difference in the DNA, but this need not necessarily manifest itself as a phenotypic difference.

The resolving power of the technique can be used for a variety of different purposes eg. to authenticate the identity of a strain grown by a yeast supply company, or to monitor a strain in a mixed strain fermentation. The technique works on colonies, yeast cake, liquid cultures and spray dried yeast. Patterns can be obtained from very small samples.

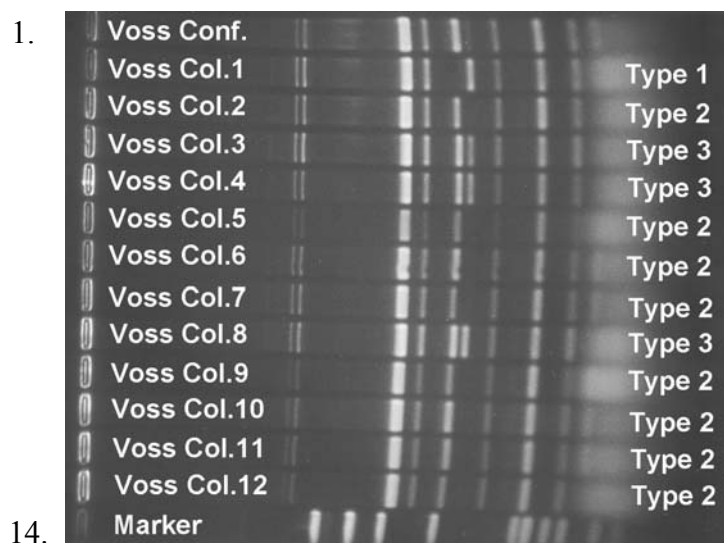
Analysis of Sigmund Gjernes Voss yeast sample:

A liquid yeast sample of the Voss yeast was provided for analysis and accession by Lars Marius Garshol. A small amount of the yeast was removed from the liquid sample, diluted in sterile water and streaked on YM agar and Rose Bengal + Antibiotic plates to provide confluent growth and single colonies for analysis.

After 3 days incubation at 25°C confluent growth and colonies on the plates were of sufficient size for analysis.

No bacteria or other contaminants were seen on the streak plates made from the Voss yeast sample.

The fourteen tracks seen on the photograph of the gel are as follows:



1. Confluent growth from Voss Sample
2. Single colony 1 from Voss Sample
3. Single colony 2 from Voss Sample
4. Single colony 3 from Voss Sample
5. Single colony 4 from Voss Sample
6. Single colony 5 from Voss Sample
7. Single colony 6 from Voss Sample
8. Single colony 7 from Voss Sample
9. Single colony 8 from Voss Sample
10. Single colony 9 from Voss Sample
11. Single colony 10 from Voss Sample
12. Single colony 11 from Voss Sample
13. Single colony 12 from Voss Sample
14. Standard Marker

Results:

Three different pattern types were seen on the gel:

Individual colony 1 has a unique pattern type. This was designated Type 1.

Individual colonies 2, 5, 6, 7, 9, 10, 11 and 12 all had the same pattern type. This was designated Type 2.

Individual colonies 3, 4 and 8 all had the same pattern type. This was designated Type 3.

The confluent growth had a pattern which was a composite of the three types seen in the individual colonies.

Conclusions:

- 1) Three different pattern types were produced. The Voss yeast sample is therefore composed of a mixture of strains.
- 2) The three different pattern Types are similar to each other, differing only by the presence of absence of a band when compared to each other. They are therefore highly likely to be related strains, either being derived from each other or sharing a common parent strain.
- 3) The confluent growth pattern was a composite of the other patterns. It is therefore likely that the 3 strain types found make up the vast majority or entirety of the composition of the Voss sample.
- 4) No contaminants, either bacterial, non-*Saccharomyces* or *Saccharomyces* unrelated to the Voss sample strain were detected.

Overall Conclusions:

Based on the analysis of the supplied Voss sample:

- 1) The sample appears to be composed entirely of a mixture of three closely related strains of yeast with Type 2 being the dominant type.
- 2) The Voss sample appears to be free from bacteria, non-*Saccharomyces* or *Saccharomyces* unrelated to the sample.

Accession and storage work:

Confluent growth and a single colony representative of each pattern type were removed from the agar plate, assigned NCYC collection numbers and stored in liquid nitrogen and by freeze drying.

Confluent growth (mixed sample) = NCYC 3995

Colony 1 / Type 1 = NCYC 3996

Colony 9 / Type 2 = NCYC 3997

Colony 8 / Type 3 = NCYC 3998