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Identity verification of trees in the 61 year old common ash (*Fraxinus excelsior*) clonal seed orchard FP202 (Birkemarken, Humlebæk) by DNA genotyping with microsatellite marker

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Preface

Denmark has a long-standing tradition for working with applied forest genetics based on seed from selected, healthy, productive and straight elite trees. Over the previous 60 years this work has implied a close cooperation between the Arboretum in Horsholm and The Tree Improvement Station in Humlebæk. The Arboretum (since 2004 a part of Forest & Landscape, University of Copenhagen) has over the years selected, developed and tested superior seed-trees, whilst The Tree Improvement Station (today a part of The National Forest & Nature Agency, Øresund) has worked with effective propagation and distribution of improved material on a national scale. Strategies as well as tangible plans are regularly discussed and coordinated. The cooperation has been a strong link between research on the one side and forestry on the other, and since 1947 the Forest and Nature Agency has established, managed and harvested seed from an extensive network of seed sources, which, as far as seed orchards are concerned, have all been established in interaction with Forest & Landscape for breeding purposes.

One of the first seed orchards in the systematic breeding work is FP202. Selection and grafting of particularly interesting ash trees was started in 1934 in the Forest Botanical Garden under C. Syrach-Larsens direction (Larsen, 1945). In 1945-46 there was a large scale propagation of 14 selected clones. Helmuth von Barner was responsible for planting 977 of these graftings at an area close to the newly started Tree Improvement Station in 1947, thereby establishing the (to our knowledge) World’s first Ash seed orchard. FP202 has since developed into one of the most important seed sources of ash in Danish forestry. In later years it has been the preferred seed source in Danish Forestry due to its origin from straight trees, but also because FP202 is expected to have the highest level of genetic diversity among the three ash seed orchards on the Danish market.

Due to the recent problems with ash dieback, special interest for the clones in FP202 has arisen. Dieback is supposedly caused by a fungus ‘Chalara fraxinea’. Though there are still many unanswered questions concerning this serious illness (Thomsen et al., 2008), it is very interesting that some clones show considerable resistance. FP202 contains both resistant and susceptible clones, and since 2007 it has become particularly interesting to study progenies from FP202 in order to increase the understanding of the genetic aspects of the disease and the option for breeding to increase resistance (Olrik et al., 2007). This work has included selective harvest from healthy clones in 2008.

Many years have passed since the first elite ash tree was selected and grafted in the 1930’ies, and there are many steps where mistakes could have occurred since. It is not possible to discern clones from each other by their appearance, and correctness of the clone identity in FP202 is therefore completely dependent on the high degree of care taken by all those involved from 1930’ies up to today. Fortunately, it is now possible to control the clone identity by DNA markers, and with this knowledge we decided to
control the clone identity of FP202, 60 years later. This report deals with the technique, results and conclusions of the analysis. In the appendix we have selected documentation concerning FP202’s establishment and treatment material which has only partly been published earlier.

Helmuth von Barner and C. Syrach-Larsen (who founded and managed the seed orchard for more than 40 years) would have been delighted by the results. There are only very few technical errors in this the (maybe) oldest ash seed orchard in the world.

Hørsholm og Humlebæk, December, 2008

Erik Dahl Kjær, Lars Graudal og Bjarne Ditlevsen
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Abstract

The *Fraxinus excelsior* clonal seed orchard FP202 was established at Birke-
marken, Humlebæk, Denmark in 1947, and is to our knowledge the first
*Fraxinus* clonal seed orchard established worldwide. 61 years after estab-
lishment we tested clonal identity of remaining ramets by application of 4
polymorphic SSR markers. FP202 consists of alternating rows of the pre-
dominant male clone V282, and 8 predominant female clones: V702, V703,
V704, V710, V791, V792, V793, V797. However, unpublished observations
suggest that V797 may have substantial contribution to the male gamete
pool, and that the gender function of V793 also is uncertain.

Among ramets labelled as V702, V703, V704, V710, V791, V792, V793,
V797 a single genotypic mismatch was observed (Tree 24-02, probably a
rootstock). Further, one V791 ramet was mistakenly labelled V282 (Tree
09-03). An important additional finding was that V702 and V703 ramets
had identical genotypes and therefore most likely originates from the same
ortet. We cannot infer from the data if it is V702 or V703 that is the correct
name. 60 of the ramets labelled V282 (male) had identical genotypes, but 5
ramets had an alternative genotype suggesting that they form an extra male
clones represented by 5 ramets scattered in the V282 rows. The origin of this
clones is unknown, but is probably introduced by mistake during the graft-
ings in 1945-46. A single ramet labelled V282 is probably a root stock.

To conclude: 115 trees (93%) had a DNA profile that fits the documentation
even though it shall be noted that V702 and V703 ramets probably form a
single clone. 8 trees deviated from the expected: 2 trees are likely root stocks,
1 tree had an incorrect label, and 5 trees form an additional clone of so-far
unknown origin.
Danish summary

Træer i aske klonfroplantagen FP202 (Birkemarken) blev undersøgt med DNA markører for at kontrollere deres klonidentitet. FP202 består (ifølge anlægsrapport og øvrig dokumentation) af rækker med (overvejende) hanlige træer (alle klon V282) som alternerer med rækker af overvejende hunlige træer (8 kloner rækkevis: V702, V703, V704, V710, V791, V792, V793, V797). Det skal dog bemærkes at Helmuth Barner i 1976 indikerer at V797 sandsynligvis er hanlig (♂?) og at V793 er tvilsom i forhold til hanlig/hunlighed (cf. Appendix 2, nedenfor), selvom begge kloner producerede en del frø i plantagens unge år (cf. Appendix 1, nedenfor).

Resultaterne af DNA analysen er sammenstillet i Figur 1 og Tabel 2. Baseret på DNA analyserne konkluderer vi, at der i hunrækkerne kun forekommer én afvigelse (vi antager der er tale om en grundstamme). Dog bemærkes at to af de hunlige kloner (V702 og V703) med stor sandsynlighed er identiske idet de har samme DNA profil. Blandt de 67 træer i hanrækker identificerede vi ét træ som ikke passede med de øvrige, men i stedet kunne identificeres som V791. 60 træer i hanrækkerne havde identisk genotype, som vi derfor antager er V282. 5 træer spredt i hanrækkerne havde identiske genotyper, som dog afvær frø fra øvrige, og vi konkluderer på den baggrund at froplantagen indeholder én ekstra klon representeret med 5 ramater. Denne klon er sandsynligvis introduceret ved en fejl under podearbejdet i 1945-46, men oprindelsen af klonen er usikker. Endelig observerede vi ét træ i hanrækkerne, hvis genotype afvær fra alle andre træer, og vi derfor antager er en grundstamme. I alt 115 ud af de 123 undersøgte træer (93%) havde således en DNA profil som passede med det forventede ifølge dokumentationen. Et enkelt træ blev af tekniske grunde ikke undersøgt og indgår derfor ikke i analyserne.

Konklusion: Froplantagen indeholder 124 træer, hvoraf 8 træer har en DNA profil som afviger fra forventningen ud fra dokumentationen: 2 træer antages at være grundstammer, 1 træ antages plantet forkert i forhold til kortet (evt. forbyttet podekvist), 5 træer udgør én ekstra klon fordelt i hanrækkerne. DNA-analysen afslørede desuden at V702 og V703 må antages at være samme klon uden det er muligt at afgøre om det er V702 eller V703 som er den korrekte betegnelse.
1. Introduction

The *Fraxinus excelsior* clonal seed orchard FP202 located at Birkemarken close to the Danish Tree Improvement Station in Humlebæk, is the second oldest existing clonal seed orchard in Denmark. Further, it is to our knowledge the first clonal *Fraxinus* seed orchard established world wise.

The seed orchard has been one of the main providers of *Fraxinus* seed to Danish Forestry since the 1960’s and is still one of the most used seed sources in Denmark. This would be sufficient reason for investing time and effort in validating the clonal identity of the trees in this old seed orchard. However, a new severe disease which was first observed in 2003, has infected the Danish ash trees dramatically, threatening the use of this important species (Thomsen *et al*., 2008). In 2007 significant variation was observed between clones in their degree of susceptibility (Olrik *et al*., 2007) and these important differences have been maintained in 2008. The findings were based on a clonal trial with 40 clones tested at two locations (CSOs). Fortunately, the old *Fraxinus* FP 202 seed orchard includes some of the best performing clones, but also some of the inferior ones. For this reason, the verification of clonal identity in the seed orchard has become increasingly important.

In 2008 it was decided to collect seed exclusively from the healthy clones in the seed orchard. Seeds on these trees are presumably sired by the healthy clone (V282), and the expectation is therefore that such seed would have increased resistance against the disease. However, in order to be able to study such parent-offspring regression as well as in order to ensure seed collection from the correct trees, it was decided in 2008 to perform a full test of clonal identity of all trees in the seed orchard.

The present survey thus has three objectives: (i) to check the clonal seed orchard for »cryptic dysfunctions« due to grafting mistakes or seed/pollen production from root stock (cf. Hansen & Kjær, 2006), (ii) ensure seed collection from correct trees during exclusive seed collection, (iii) to facilitate precise comparison of parent-offspring regression in health and thereby allow studies on heritability and genetic background for the apparent resistance (to be studied in 2009).

The present report documents the applied methods and results of this survey with the three above objectives.
2. The Seed orchard

Establishment and management of the seed orchards are described in some detail in old reports and other written material mainly authored by Helmuth Barner, the director of the Tree Improvement station from its establishment in 1947 until 1988. Two key reports are reproduced in appendix 1 and 2, from where the below description has been extracted:

The seed orchard was established with graftings in 1947 on agricultural land. A total of 14 clones were included, 1 predominantly male clone and 13 predominantly female clones (Appendix 1). Brief description of plus trees and grafting work are included in Appendix 3, and pictures of the majority of the plus trees are included in Appendix 1. The establishment phase included mechanical weeding. Pruning of branches was applied in 1953 and 1954 to increase seed set, but with very limited effect. Application of fertiliser and removal of grass did increase the health and growth of the seed orchard trees, but did not lead to substantial seed yield (Appendix 1).

5 of the female clones were removed in 1961, leaving only 8 predominantly females (V702, V703, V704, V710, V791, V792, V793, V797), and one predominantly male (V282). However, in 1976, Helmut Barner questions if V793 and V797 are truly predominantly females (cf. Appendix 2), and these 2 clones may therefore also be important pollen donors in the Seed orchard. Further information on the selection and propagation of the individual clones are included in Appendix 3.
3. Methods

**Sampling**

Leaf material was collected on September 10th 2008. Two leaves were collected from each tree throughout the seed orchard. Each sample was labelled according to their position on the map (Figure 1). In total, 67 samples were collected from the male rows (presumed V282) and 56 samples from the female rows (presumed V702, V703, V704, V710, V791, V792, V793, V797). The material was stored at -20 °C until DNA extraction.

**Genotyping**

15-20 mg leaf tissue per individual was treated with liquid nitrogen and ground on a bead mill without any prior preparation. DNA extraction was carried out with the DNeasy 96 Plant Kit from QIAGEN following the manufactures protocol for frozen material.

The DNA-extractions were kept undiluted for the polymerase chain reaction. Eleven primer pairs developed for *Fraxinus excelsior* were initially tested. Ten of these (FEMSATL1, 2, 4, 5, 8, 10, 11, 12, 16, 19) were all developed by Lefort *et al.* (1999). FEMSATL12 has later been modified by Gerard *et al.* (2006) and it was the modified version that we tested. The last primer pair (M2-30) was developed by Brachet *et al.* (1999). Four polymorphic, interpretable microsatellite loci were identified and used for further genotyping. The chosen primer pairs were FEMSATL11, FEMSATL12, FEMSATL16, FEMSATL19.

PCR reactions were carried out using the Qiagen Multiplex PCR kit according to the manufacturer’s instructions except that the reaction volumes were scaled down to 15μl. PCR-amplifications were completed on Perkin Elmer Thermo cyclers (models 9700 and 2700) under the following conditions: an initial denaturation step of 15 min at 95 °C, 30 cycles of denaturation at 94 °C for 30 s, annealing at 57 °C for 90 s and extension at 72 °C for 60 s, and a final extension step at 60 °C for 30 min. Each amplified product was diluted with 30μl H₂O and visualized with an ABI3130xl sequencer from Applied Biosystems.
4. Results

In Table 1 we see that the amplification of the 4 microsatellite loci in total resulted in 35 alleles. This level of polymorphism was highly sufficient to distinguish between the genotyped clones.

Table 1. Number of alleles per microsatellite locus. The alleles are given in base pairs

<table>
<thead>
<tr>
<th></th>
<th>Femsatl11</th>
<th>Femsatl12</th>
<th>Femsatl16</th>
<th>Femsatl19</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>183</td>
<td>172</td>
<td>186</td>
<td>174</td>
</tr>
<tr>
<td>2</td>
<td>185</td>
<td>174</td>
<td>188</td>
<td>176</td>
</tr>
<tr>
<td>3</td>
<td>191</td>
<td>176</td>
<td>196</td>
<td>180</td>
</tr>
<tr>
<td>4</td>
<td>193</td>
<td>178</td>
<td>200</td>
<td>182</td>
</tr>
<tr>
<td>5</td>
<td>197</td>
<td>190</td>
<td>186</td>
<td></td>
</tr>
<tr>
<td>6</td>
<td>201</td>
<td>196</td>
<td></td>
<td>188</td>
</tr>
<tr>
<td>7</td>
<td>203</td>
<td>198</td>
<td></td>
<td>190</td>
</tr>
<tr>
<td>8</td>
<td>205</td>
<td>200</td>
<td></td>
<td>192</td>
</tr>
<tr>
<td>9</td>
<td>213</td>
<td>204</td>
<td></td>
<td>194</td>
</tr>
<tr>
<td>10</td>
<td></td>
<td>206</td>
<td></td>
<td>198</td>
</tr>
<tr>
<td>11</td>
<td></td>
<td>211</td>
<td></td>
<td>202</td>
</tr>
</tbody>
</table>

Of the 67 examined male trees (labelled V282) it turned out that 60 ramets had identical genotype (recognized as V282, Table 2). The remaining seven were of a different genotype. One (position 0905) turned out to have same genotype as V791 and may thus be a wrong grafting. One was not recognized elsewhere (position 0307) and may be a root stock. The last 5 (positions 0107, 0909, 1505, 1903) all had the same genotype (see Table 2) that did not match with any of the others.

Among the female rows, only 1 tree (labelled V702, position 2402) did not resemble any other genotype and may thus be a root stock. However, very interestingly trees labelled V702 and V703 turned out to have identical genotype, and it is therefore very likely that they originate from the same ortet (= the same clone).
Figure 1. Positions of trees with deviating genotypes compared to documentation. Marked in red: »Extra male« = five trees with identical genotypes that are not identical to the V282 genotype. The two squares show the trees with no match. V702 and V703 turned out to have identical genotypes.
Table 2. Summary of the genotypes of the clones of *Fraxinus excelsior* from Birkemarken (seed orchard FP202). Alleles are given in base pair lengths at each of the 4 loci.

<table>
<thead>
<tr>
<th>Clone ID</th>
<th>Femsat11</th>
<th>Femsat12</th>
<th>Femsat16</th>
<th>Femsat19</th>
</tr>
</thead>
<tbody>
<tr>
<td>V282</td>
<td>183</td>
<td>201</td>
<td>174</td>
<td>174</td>
</tr>
<tr>
<td>V702</td>
<td>191</td>
<td>205</td>
<td>176</td>
<td>196</td>
</tr>
<tr>
<td>V703</td>
<td>191</td>
<td>205</td>
<td>176</td>
<td>196</td>
</tr>
<tr>
<td>V704</td>
<td>193</td>
<td>213</td>
<td>190</td>
<td>206</td>
</tr>
<tr>
<td>V710</td>
<td>183</td>
<td>203</td>
<td>172</td>
<td>190</td>
</tr>
<tr>
<td>V791</td>
<td>185</td>
<td>191</td>
<td>174</td>
<td>178</td>
</tr>
<tr>
<td>V792</td>
<td>183</td>
<td>191</td>
<td>174</td>
<td>174</td>
</tr>
<tr>
<td>V793</td>
<td>183</td>
<td>191</td>
<td>200</td>
<td>204</td>
</tr>
<tr>
<td>V797</td>
<td>197</td>
<td>205</td>
<td>190</td>
<td>200</td>
</tr>
<tr>
<td>Extra male</td>
<td>183</td>
<td>191</td>
<td>174</td>
<td>190</td>
</tr>
<tr>
<td>(0107, 0901, 0909, 1505, 1903)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Male with no</td>
<td>193</td>
<td>205</td>
<td>174</td>
<td>211</td>
</tr>
<tr>
<td>match (0307)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Female with no</td>
<td>197</td>
<td>201</td>
<td>190</td>
<td>198</td>
</tr>
<tr>
<td>match (2402)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Please note that V702 and V703 have identical genotypes and it is therefore highly likely that it is indeed the same clone.
5. Conclusion

In conclusion, the seed orchard corresponded to 7 clones in female rows and 2 clones in the male rows rather than 8 clones in female rows and 1 clone in male rows. Besides this, there were only few unexplained deviations (1 assumed wrong/misplaced grafting, and 2 likely root stocks). According to old records, 2 of the clones in the female rows may produce significant amount of pollen and the pollen production id therefore likely to be distributed on more than one clone.

The identity of the »extra male« remains unknown. We have genotyped 40 clones deployed in different Danish seed orchards, but none of these match (Nielsen, unpublished results). A screening of old clonal archives with Fraxinus may reveal the true identity of this »extra male«, but this will probably not be an easy matter. Studies of segregation in terms of health of progenies from FP202 will be performed in 2008-2009, and here we will look for progenies sired by this »extra male«. Based on this parental analysis we expect to be able to determine the health status (in term of breeding value) of the »extra male«. This will only be possible if the clone has been sufficiently male fertile to sire a fair number of progenies in our progeny sample. Based on the results it will be possible to recommend if the clone should be removed from the seed orchard or maintained.
6. References

Rapid identification of microsatellite loci using 5’ anchored PCR in the com-
mon ash (Fraxinus excelsior). Molecular Ecology 8, 157–168.
Temporal cline in a hybrid clone population between Fraxinus excelsior L. and
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seed orchard reveals dysfunctions. Canadian Journal of Forest Research 36,
1054–1058.
Larsen, C.S. 1945.
Identification and characterisation of microsatellite loci in ash (Fraxinus excelsior L)
and their conservation in the olive family (Oleaceae). Molecular Ecology 8, 1088–1091
Klonforskelle i angreb af asketoptørre. SKOVEN 39, 522–525
FP 202, December 2008. Pruning has been applied to increase fruiting and ease seed collection (cf. Annex 1 below). Many trees therefore have low twigs. However, today the clonal identity is correct on most trees and the DNA analysis only identifies two trees to be likely root stock. This result proves careful pruning and removal of root stock sprouts in the seed orchard.
Appendix 1: Description of the establishment of FP202

Source: Selected part of the unpublished document ‘Frøhaver Ask’, which is part of the old files at the Arboretum. The file is undated and the authorship is not stated. We believe it was written by Helmuth Barner, the former director of The Danish National Tree Improvement Station, who was responsible for the establishment and management of the seed orchard.

We are not sure about the date. The document refers to the flowering in 1960, and is likely to be prior to 1961, because it does not refer to the thinning of the clonal seed orchard that was performed in that year according to documents from 1976 (appendix 2 below). The document include numbers of seed harvested up till 1966/67, but these figures may have been added later as no references are given to flowering after 1960. We therefore assume that the document dates around autumn 1960.
Planteavlstationen
Birkemarke
Areal: 1.5 ha.

Ansk
Prehave 202.

Formål: Produktion af askesø.  
Isolering: Isolering fra omgivende ask ret god, ca. 500 m gennem skov til nærmeste askesøbekøbeling, der er gammel og næppe blomstrer mere.

Arvemateriale: Som huntrer indgår i plantagen følgende, der er udvalgt af dr. Syrach Larsen:

<table>
<thead>
<tr>
<th>V.</th>
<th>Afd.</th>
<th>Træ nr.</th>
<th>Træer</th>
<th>Årstal</th>
</tr>
</thead>
<tbody>
<tr>
<td>702</td>
<td>Nørreskov</td>
<td>12</td>
<td>1</td>
<td>1946</td>
</tr>
<tr>
<td>703</td>
<td>Nørreskov</td>
<td>1</td>
<td>2</td>
<td>1945</td>
</tr>
<tr>
<td>704</td>
<td>Nørreskov</td>
<td>30</td>
<td>40</td>
<td>1946</td>
</tr>
<tr>
<td>710</td>
<td>Høvsgård</td>
<td>5</td>
<td>40</td>
<td>?</td>
</tr>
<tr>
<td>793</td>
<td>St. Bøgeskov</td>
<td>63</td>
<td>80</td>
<td>1945</td>
</tr>
<tr>
<td>793</td>
<td>St. Bøgeskov</td>
<td>67</td>
<td>20</td>
<td>1945</td>
</tr>
<tr>
<td>793</td>
<td>Ll. Bøgeskov</td>
<td>82</td>
<td>60</td>
<td>1946</td>
</tr>
<tr>
<td>797</td>
<td>Stenævang</td>
<td>274</td>
<td>40</td>
<td>1945</td>
</tr>
<tr>
<td>869</td>
<td>Dalby Skov</td>
<td>23</td>
<td>4</td>
<td>1945</td>
</tr>
<tr>
<td>894</td>
<td>Greveaskoven</td>
<td>90</td>
<td>4</td>
<td>1945</td>
</tr>
<tr>
<td>895</td>
<td>Greveaskoven</td>
<td>-</td>
<td>2</td>
<td>1945</td>
</tr>
<tr>
<td>895</td>
<td>Greveaskoven</td>
<td>-</td>
<td>112</td>
<td></td>
</tr>
<tr>
<td>896</td>
<td>Greveaskoven</td>
<td>-</td>
<td>5</td>
<td>1945</td>
</tr>
<tr>
<td>899</td>
<td>Kimmelslev, Heskel</td>
<td>11</td>
<td>2</td>
<td>1945</td>
</tr>
</tbody>
</table>

Som han er anvendt V. 202 Stønderup Nørreskov.

Fril. F.O. Træ nr. 44. 500 stk. (til og med 5 rk. 10. plt. p. 46 (derfra og til og med 34 rk. 16. plt.) fra (ok.47, resten ok. 44 ayd)

Plantagen er anlagt i foråret 1947 i den østligste del af Birkemarke, V. 202 indgår med 2 rækker i hver anden dobbelt række, huntrerne indgår derimellem som vist på skitser.

Alt materiale er leveret af Arboretum i Hersholt.

Planteafstand 4 x 4 m.

Plantagenes udvikling: Plantagen blev anlagt på gl. ajerjord, der lige efter overtagelsen blev behandlet med harvning, hvorefter askene blev udplantet. De første 3 år blev der til stædighed venhølt med harvning, hvorefter man indskrænkede sig til at holde grøset nede med slaming.

For at prøve dels at påvirke frøsmningen, dels at lette plukningen, klippede man første gang i vinteren 1953/54 i rækkerne mellem huntrer, gennem nind på hveranden ask i hver række (1 for bundet). Dette blev gentaget i 1954/55 med de samme træer. - Intet udslag!
fot. Juli 1956, Fl.
Efter at man i nogle år havde holdt arealmet med gres, viste det sig, at aksene hæmmedes i udviklingen. Slidene blev gullige, og vejksten var særlig på midterarealet ringe. I oktober 1957 fræsede man derfor mellem alle akserrækkerne (4 træk mellem 2 rækker aek). Følgende rækker mellemrum blev dog ikke fræsede:

25 - 26 = V. 262
33 - 34 = V. 262

Idet rækkerne regnes fra nord.

Umiddelbart herefter blev på tvers af rækkerne givet

7 rækker aek = 225 kg svovlsur ammoniak ialt
6 - - = intet
7 - - = 225 kg kalksalpeter ialt

I foråret 1958 blev følgende tilført:

7 rækker aek = 250 kg svovlsur ammoniak ialt
6 - - = intet
7 - - = 250 kg kalksalpeter ialt

Allerede tidligt på sommeren 1958 sås klart udslag for gødskningen. Idet 2x7 gøde rækker fik mørkegrønne, mundde blade. Der konstateredes ikke forskel på de 2 gødningsartere virkning.

Foråret 1959 gødedes hele arealmet med 375 kg svovlsur am. og senere med 375 kg. Arealmet blev i 1959 holdt med fremsning og er i 1960 blevet havret. Gødskning + fremsning gav klart positivt udslag m.h.t. vækst.

I 1950/59 blev foregået en kraftig beæskering af hveranden 1 træ. Virkningen var, bedstent sommeren 1960, ikke særlig gunstig. Der har som frygtet udviklet sig kraftige vannir på de beækkerne træer og blomstringen forår 1959 var ikke bedre på de beækkerne træer end på de andre træer.

1960 synes at være godt blomstringsår for aek. Blomstringen i frehaveren er dog beskeden og i lighed med de 2 tidligere blomstringsår, er der flest blonster i de laveste dele mod S. og N. Der er dog en nogenlunde god blomstring på 9 rækkerne også på midterstykket, hvilket tyder på, at det ikke alene er de ydre forhold, der hindrer de klonerne i blomstring på midterstykket.
Amk.
Søre I, Broby Vesterkov, afd. 63 a.
V. 791.
Fot. den 29/1 -1954.

Amk.
Tåninge, Horre skov, afd. 5.
Træ mrk. 3 gule platt. V.7loc.
Fot.

Amk.
V. 799.
Fot. den 29/1 -1945.
Ask.
Bregentved, Sorte lad afd.112.
V.895.
foto den 23/1 - 1954.

Ask.
Svenstrup, Kimmeralov Hessel afd.11
V.898.
foto 6.22/1 - 1954.

Ask.
Bregentved, Jubilæumsseksen.
V.896.
foto 8.22/1 - 1954.
Ank.
Stenderup Nørrebo, afd. 12.
træ nr. 1, V. 702.

Ank.
Boller Nørrebo, afd. 30.
træ (gul ring), V. 704.
fot. den 30/1-44.

Ank.
Stenderup Nørrebo, afd. 30.
træ nr. 2, V. 703.
Appendix 2: Description of FP202, 1976


PROVENIENSMEDDELELSE NR. 7
STAMBOGEBLAD
for
FP202BIRKEMARKEN FP202

1. Identification
1. Træart: Åsk, Fraxinus excelsior
2. Læbenummer: 202
4. Ansvarlig tilgivnings-Planteavlslstationen
6. Areal: 1.5 ha
7. Kort: A 2630

1. Fraets anvendelsesområde: Ingen særlige begrænsninger inden for danske askelokaliteter.

3. Udbygning
1. Komponenter | Antal ved anlæg | Antal fjernet | Formeringsår | Udplantningsår | Frekæst vigtigt indtil
Kloner | 14 | 1951 | 5 | 1945-46 | 1947 | 1/1-76 1241
Familier | 0

3. Plantningsafstand: 4 x 4 m
4. Isolering: Enkelte små askegrupper 200-300 m N.V. for plantagen, enkelte aske ca. 450 m gennem skov N. og NV for plantagen, samt 2 små askebevoksering ca 350-600 m gennem skov NV for plantagen.

4. Grundmateriale
2. Udvalgsejendomme. Efter kun for at få hønlige og hønlige kloner samt efter ræhed og stammekvalitet.
3. Udvalgsstræke: Vanskeligt at vurdere.
# 5. Komponenternes og deres afstemning.

<table>
<thead>
<tr>
<th>Betegnelse</th>
<th>Køn</th>
<th>Oprindelser</th>
<th>Herkomst</th>
<th>Antal træer i plantagen 1945</th>
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<tr>
<td>V 702 træ 1</td>
<td>♂</td>
<td>Formentlig dansk lokalrace</td>
<td>Stenderup, Nørreskov afd. 12</td>
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<td>20</td>
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<td>V 703 - 2</td>
<td>♀</td>
<td>lokalrace</td>
<td>Stenderup, Nørreskov afd. 10</td>
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<td>20</td>
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<tr>
<td>V 704</td>
<td>♂</td>
<td>Boller, Neder skov afd. 30</td>
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<td>40</td>
<td>10</td>
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<td>V 710</td>
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<td>Tásinge, Hor semovk afd. 5</td>
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<td>Søs II, L2. Bgeskov afd. 82a</td>
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<td>V 797, træ 1</td>
<td>♀</td>
<td>Harsholm, Stasevang afd. 274</td>
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<td>V 869</td>
<td>♀</td>
<td>Svenstrup, Dalby afd. 23a</td>
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<td>Bregentved, Gre vinde afd. 9c</td>
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<td>V 895</td>
<td>♂</td>
<td>-</td>
<td>-</td>
<td>112</td>
<td>2</td>
</tr>
<tr>
<td>V 896</td>
<td>♀</td>
<td>-</td>
<td>-</td>
<td>Bregentved &quot;Jubilmumsask&quot;</td>
<td>5</td>
</tr>
<tr>
<td>V 898</td>
<td>♀</td>
<td>Svenstrup, Kimmerslev afd. 11</td>
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<td>2</td>
<td>0</td>
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<tr>
<td>V 282 træ 44</td>
<td>♂</td>
<td>Stenderup, Nørreskov afd. 12</td>
<td></td>
<td>500</td>
<td>122</td>
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<tr>
<td>Ialt</td>
<td></td>
<td></td>
<td></td>
<td>977</td>
<td>241</td>
</tr>
</tbody>
</table>

# 6. Forventninger til fremplantagens afkom.

1. Udgangspopulationens niveau: Bevokkninger over middel.
2. Puntnærvælgtes forventede effekt: Usikker i en så stærkt karplotvirkelig art.
3. Forventet heterosisseffekt: Ingen-ringe, event. provensa/hybrid-effekt.
4. Forventet kombinationseffekt: ?
Denne proveniencesseddelelse aflæser den tidligere udsendte proveniencesseddelelse af 7 af september 1963, der heddes udtaget af samlingen.
Appendix 3: Selection and propagation of clones in FP202 (1938-1944)

Source: Scanned copies of original registrations made during 1938-1944. Unpublished records kept at Forest and Landscape, Faculty of Life Sciences, University of Copenhagen.

V282-1
Første 1941: Bedst 298 stk. af stærke Medicale i plantet brille foran Livet.

80 Svinger. Jernangang fætter 1941 af Nykøbing, 195 stk. ikke blandt de

12/11 480 stk. vældt med middelhøj 66,8 cm, stiveste højde 160 cm.

2/11 41. Planen afinheritDoc og sortiment.

203 stk. smøkket, redde det udendørs

150 - med sammert Svinj - - -

35 - til redskaberet

148 - til redskaberet

15 - mossel

+ 14 - kassette for råhaver (fremad i bækkøgen af Nykøbing Maribo)
26/11-44: Material handlet af E. Magsen, der beskriver Træd:
Totalkjærde 29.3 m, Øaen i kjærde 16.0 m. Øens. o.65 m
(lyd: Sdr. Læsøs kaer). Træet har vandt værdi
og rige klippe bestand af frø, der er derom
om den tidseje og er kommet vej i
Skr. : Magsens henvisning S. 2.
14-44: 1000 cm² skrå = 76.25 g.

7/3-44: Potet i ár 12 stk.
1/7-44: Ris 10 stk.

27/5-44: Potet fra kommetjede 32 stk.
10/7-44: Ris 12 stk.

26/45: Potet i åre afk 20 stk.
2/47: Ris 13 stk.

V703

26/44: Material humid till 1. Maajis, der skriver:

"Tolthøjde 24.2 m, Aethøjde 14.7 m, Øjenhøjde 4.5 cm
Ike. Lassen medarbejder. Også dette fra hav fremad
følgt yderligere Tøj, men her var ikke ret mange
hjælpe og det lås meget lidt, saa det mange faldt
af, naar gennem faldt til jorden."

Skriver: Maajis medarbejder S. E.

4/44: 1000 Kar Vage: 76.25 q

4/5-44: Fodet i havo 13 skt.
4/44: Rast 10 skt.

24/5-44: Fodet fra Varemølle 30 skt.
14/7-44: Rast 21 skt.

24/6-45: Fodet i Ærøskov 20 skt.
7/6-47: Rast 3 skt.

Ind [nt, 60]
3. 44: Anlægget hændel af E. Magini, der skrevet:

"Totalt 23,2 m. Aftårige 18 m. Buthårige 15 m.
Anlæg. 1,8 m. Høj 116 cm. Et meget smukt Vær. Stam-
mens mestre fuldstændig ret, hæren var, formelt, men
ikke ved sten. Kairt meget lidt sit, som hænder tildelt
jævnt jordet om Trækl.

...... Trækl var ret
Fremmover, men med sid stammer var en del engag-
formet af stens...."

Skijs: Foto i Magini's Register 18. 44.

$90. 44: 1000 km. Vægt: 98 g.

$9. 44: Voldt i hus 12 stk.

$7. 44: Alle i lin.

$7. 44: Voldt p. Førerkystenlig 29 stk.

$7. 44: Huk 27 stk.

$7. 44: Fra til Carl Kolding og ulige plantes både i nabøen.

$7. 44: (i de øvrige) nærmere 1 g med mindre mellemstørrelse 0,9

$7. 44: (i en mindre) 0,7 og 0,8, men kan nu se, at han

gar nøglet Barber, bl.a. mere Inde af 0,3

Bogstavelige plantes har været isoleret i godt lid.
V704 (2)

... og undersøgte sig lyver de Frie er de
viten schizotypi og deres dans med Bevægning.

2.6.45: Døde i Søgade 20. Stk.

Indført 1950
V. 710.

Q. Sek.

Agr. 5

Store Skor v. Tangeby. Vedkommende D.

143-44: Materialer hentet af S. Magin, den skues:
1. Tølthjælpe 21 m. 2. Tølthjælpe ... 3. Tølthjælpe 12.3 m.
Afg. 78. cm. Skallen ikke hættet, men indes
Fremlæg. Hanen selveget, jævnt og rigtigt forret
med Brø. Trael støvet i en bevodning med mange
mængde småt. dør, som dog først bliver tages ad van-
ne Trael. Enkelt Trael med hjælpe af fællesgiv-
grammen. Høyerer结晶er af ca. 300. Pud-
Væske og instaner. Fjerke af de nedskaffede jern.

14-44: 1000 Rom Vajt: 100.6 g

13-44: Podd i hus 12 stk.
19-44: Podd 12 stk.
-44: --- 10 ---

245-44: Podd fra Stemmehaft 38 stk.
1917-44: Podd 32 stk.

Træn 45: Podd fra Stemmehaft 15 stk. (omrødning)

246-45: 20 stk. podd i åndag.
76-47: Podd 15 stk.
V791

V. 791.

All, Srty Veletischer, 1 Soro, Aug. 63 ar

25/54: Proof & This 10 Sec.
4/54: Trade & Live

Ind str 1/80
31. 9. 93.
Rød, "Frisk"
Lille Bogser afk. 82 é. Paaf. FK é.
Jubilæum g. fød. 23. 12. 1845.

24/5-44: Poter paa Vemundtofte 34 stk.
17-11-44: Rent 24 stk.

24/5 45: Pakket i bagvogn 20 thb.
7/6 47: Rent 16 thb.

indford/51
29. IV. 44: Order: Here 13 Lk.
1/4 44: Rest 11 Lk.

29/5 44: Order pair Vermeschip 20 Lk.
10/7 44: Rest 12 Lk.

V 797
<table>
<thead>
<tr>
<th>No.</th>
<th>Year</th>
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<tr>
<td>1</td>
<td>2004</td>
<td>Experiences with web-based teaching in forestry</td>
</tr>
<tr>
<td>2</td>
<td>2004</td>
<td>Distribution of tree seed and seedlings</td>
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<tr>
<td>3</td>
<td>2004</td>
<td>Identifying forest-livelihood research priorities in Mozambique</td>
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<tr>
<td>4</td>
<td>2004</td>
<td>Breeding for die-back resistant <em>Dalbergia sissoo</em> in Nepal</td>
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<tr>
<td>5</td>
<td>2005</td>
<td>Farmers’ planting practices in Burkina Faso</td>
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<td>6</td>
<td>2005</td>
<td>Cocoa agroforests in West Africa</td>
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<tr>
<td>7</td>
<td>2005</td>
<td>Observations on timing and abundance of flowering and fruiting of woody plants</td>
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<tr>
<td>8</td>
<td>2005</td>
<td>Tree seed in Malawi</td>
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<td>9</td>
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<td>Commercial distribution of tree seed in small bags</td>
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<td>2005</td>
<td>Using soft systems methodology to develop a mango forest management and planning decision support system in a buffer zone</td>
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<td>18</td>
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<td>A review of forest economics research in Bolivia</td>
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<td>Proceedings of a workshop on agroforestry tree seeds for farmers</td>
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<td>Protocol for establishment of trials with Baobab and Tamarind within the SAFRUIT project</td>
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<td>Evaluation of an international series of Pinus kesiya provenance trials for adaptive, growth and wood quality traits</td>
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<td>Larch wood – a literature review</td>
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<td>The potential of larch wood for exterior use – Report from a joint Nordic research project</td>
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<td>A floral and faunal biodiversity assessment of Prey Long</td>
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<td>Proceedings of the 8th International Christmas Tree Research &amp; Extension Conference</td>
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<td>Innovation Systems and Rural Development</td>
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