



## **Interpretation of Oiled Feather Data from the *M/V New Carissa* Spill**

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*M/V New Carissa* photo from Ship Structure Committee website

# Interpretation of Oiled Feather Data from the *M/V New Carissa* Spill

James R. Payne and William B. Driskell

## Task Description

As a follow-up to earlier Natural Resource Damage Assessment (NRDA) activities following the 1999 *M/V New Carissa* oil spill, 61 archived feather samples were analyzed in a blind study to determine whether oil extracted from feathers matched known source oils from the *New Carissa*. The samples were analyzed by the Battelle New England Laboratory in May 2003, and data were distributed to Trustees' and Principal Parties' experts. This report discusses the methods and conclusions of the Trustees contractor, Payne Environmental Consultants, Incorporated (PECI) while the data were still under blind status.

## Methods

The *M/V New Carissa* spill has produced a challenging data set for spill assessors. As explained in earlier reports (Payne and Driskell 1999, 2000, 2001), the *New Carissa* spill comprises at least 5 oil sources in various states of mixing and weathering, i.e., the signature is very complex. Fortunately, the earlier efforts allowed us to extend the prior knowledge of *New Carissa* spill characteristics to the current oiled-feather dataset. The data were comparable to earlier analyses reporting polycyclic aromatic hydrocarbon (PAH), saturate aliphatic hydrocarbon (SHC), and sterane/triterpane (S/T) analytes. Table 1 lists the target compounds for each of these three analyte groups and presents the abbreviations used in all histogram plots and other figures in this report. For the feather samples, the data quality was extremely good (excellent surrogate recoveries), and the fingerprint characterization data for the source oils were directly comparable with previous chemical analyses completed at A.D. Little (ADL).

## Graphic Analyses

With blind samples (no spatial or temporal information), interpretation was primarily limited to comparisons of histogram-plot analyte signatures. To this purpose, an Excel data graphing utility was developed to permit easy comparison of individual samples against multiple individual and mixed source oils.

It was immediately obvious that the 19 reference oils included mixtures that overlapped in their compositions and that there were significantly fewer actual sources. This was confirmed by looking at the classic double-ratio plots of refractory analytes, namely, alkylated dibenzothiophenes to alkylated phenanthrenes and alkylated dibenzothiophenes to alkylated chrysenes (Brown et al. 1980; Overton et al. 1981; Boehm et al. 1989; Sauer and Boehm 1991; Brown and Boehm 1993; Page et al. 1993; Page et al. 1995; Douglas et al. 1996).

**Table 1 List of target analytes and abbreviations.**

Polycyclic Aromatic Hydrocarbons (PAH)		Saturated Aliphatic Hydrocarbons (SHC)		Steranes/Triterpanes (St/T)	
N	Naphthalene	C8	Octane	T4-C23	T4-C23 Diterpane
N1	C1-Naphthalenes	C9	Nonane	T9-C29	T9-C29 Tricycliterpane
N2	C2-Naphthalenes	C10	Decane	T10-C29	T10-C29 Tricycliterpane
N3	C3-Naphthalenes	C11	Undecane	T11-Tri	T11-Trisnorhopane (TS)
N4	C4-Naphthalenes	C12	Dodecane	T12-Tri	T12-Trisnorhopane (TM)
BI	Biphenyl	C13	Tridecane	T15-Nor	T15-Norhopane
AC	Acenaphthylene	1380	Isoprenoid 1380	T18-Ole	T18-Oleanane
AE	Acenaphthene	C14	Tetradecane	T19-Hop	T19-Hopane
F	Fluorene	1470	Isoprenoid 1470	T21-Hom	T21-Homohopane
F1	C1-Fluorenes	C15	Pentadecane	T22-Hom	T22-Homohopane
F2	C2-Fluorenes	C16	Hexadecane	S4-Diac	S4-Diacholestane
F3	C3-Fluorenes	1650	Isoprenoid 1650	S5-Diac	S5-Diacholestane
D	Dibenzothiophene	C17	Heptadecane	S24-Met	S24-Methylcholestane
D1	C1-Dibenzothiophenes	Pristane	Pristane	S25-Eth	S25-Ethylcholestane
D2	C2-Dibenzothiophenes	C18	Octadecane	S28-Eth	S28-Ethylcholestane
D3	C3-Dibenzothiophenes	Phytane	Phytane		
D4	C4-Dibenzothiophenes	C19	Nonadecane		
P	Phenanthrene	C20	Eicosane		
A	Anthracene	C21	Heneicosane		
P/A1	C1-Phenanthrenes/Anthracenes	C22	Docosane		
P/A2	C2-Phenanthrenes/Anthracenes	C23	Tricosane		
P/A3	C3-Phenanthrenes/Anthracenes	C24	Tetracosane		
P/A4	C4-Phenanthrenes/Anthracenes	C25	Pentacosane		
FL	Fluoranthene	C26	Hexacosane		
PY	Pyrene	C27	Heptacosane		
F/P1	C1-Fluoranthenes/Pyrenes	C28	Octacosane		
F/P2	C2-Fluoranthenes/Pyrenes	C29	Nonacosane		
F/P3	C3-Fluoranthenes/Pyrenes	C30	Triacontane		
BA	Benz[a]anthracene	C31	Hentriacontane		
C	Chrysene	C32	Dotriacontane		
C1	C1-Chrysenes	C33	Tritriacontane		
C2	C2-Chrysenes	C34	Tetratriacontane		
C3	C3-Chrysenes	C35	Pentatriacontane		
C4	C4-Chrysenes	C36	Hexatriacontane		
BB	Benzo[b]fluoranthene	C37	Heptatriacontane		
BK	Benzo[k]fluoranthene	C38	Octatriacontane		
BEP	Benzo[e]pyrene	C39	Nonatriacontane		
BAP	Benzo[a]pyrene	C40	Tetracontane		
PER	Perylene				
IP	Indeno[1,2,3-c,d]pyrene				
DA	Dibenz[a,h]anthracene				
BP	Benzo[g,h,i]perylene				

## Blend Assessment

Subsequently, another Excel application was developed to assess the blends of the 5 primary oil sources in each of the identified *New Carissa* impacted samples. This application included programming a linear optimizing routine to fit the blend of different source oils to each sample's unique signatures. Although the routine was designed to separately optimize each of the three analyte groups (PAH, SHC, and S/T), in practice, the PAH and S/T optimizations proved most relevant to the source identification process. Performing the actual calculations was the Excel add-in procedure, Solver.XLA, but the resulting mathematical fit was never considered the "final answer." In all cases, the final optimization results were scrutinized to assess their validity based upon expert knowledge of source oil composition, mass balance considerations, and weathering patterns.

One of the issues in matching a blend of source oils to a given sample is to decide which of the analytes in a given group is most relevant for comparisons, i.e., choose a matching model that works. For example, many of the lower-molecular-weight PAH compounds (naphthalenes (N) and fluorenes (F)) weather quickly. Furthermore, the parent PAH within each analyte group is more subject to loss by evaporation and dissolution in water compared to the higher-alkylated homologues. For our purposes, the more persistent alkylated homologues of dibenzothiophenes (DBT), phenanthrene/anthracenes (P/A), fluoranthene/pyrenes (F/P) and chrysenes (C) were most relevant. For a first-pass blending estimate, the optimization routine simply tries to match ratios of the group totals between the sample and reference blend, i.e., adjusting the blend to minimize the error in the fit of the sum of DBTs (C-1 to C-4) versus the sum of P/As (C-1 to C-4), etc., for the four groups. In the final step of interpretation after the blends are constructed, the proportions of the individual homologues within their respective groups are considered equally or even more important than getting a "tight-fitting" blend. Fortunately, for this data set, the blending estimate was remarkably good at fitting a blend to the actual data. Verification of the blend was validated by the visual similarity in comparing the resulting histograms of blend versus sample. A statistical goodness-of-fit index, the optimal fit value, was also generated.

For the SHC analytes, the optimization results were not as useful. Fitting was based upon the relative proportions of the isoprenoids 1370, 1460, pristine, phytane and the C<sub>21</sub>-C<sub>30</sub> n-alkanes. But often, the visual modality of the histogram was just as useful. In contrast, the sterane/triterpane comparisons used the full suite of analytes. Here the optimal fit routine was very useful because plot modality was not obvious.

## **Results**

### Sources and Blends

The first task was to compare the sources: 19 potential source oils were analyzed relative to the testament samples (provided by the ship's engineer from aliquots collected at the time of fueling) and spill mixtures obtained adjacent to the *New Carissa* at the time of the

spill. Figures 1 and 2 present characteristic histogram plots for the PAH, SHC, and S/T analyte groups for the five major ship sources plus one of the blends collected from the beach adjacent to the vessel on February 14, 1999. All concentrations are in  $\mu\text{g/g}$  (parts per million [ppm]) for total polycyclic aromatic hydrocarbon (TPAH), total saturated hydrocarbons (TSHC), and total steranes and triterpanes (TSSt) presented above each plot. The units for all concentration (y-axis) values are also  $\mu\text{g/g}$ . Abbreviations for target analytes are presented in Table 1.

Fortunately, the PAH, SHC, and S/T distributions of the testament samples are highly disparate which simplifies the identification process. The BFO 280 cSt 1/19 sample is predominated by alkylated naphthalenes (N1-N4). The PAH histogram for the MFO 280 11/28 sample is lighter in naphthalenes compared to the other PAH in the sample, and the alkylated dibenzothiophenes (D2 and D3) are the most predominant group. It is also important to note, however, that the TPAH concentration for the MFO 280 11/28 is only 5,285 ppm (0.53%), while the TPAH concentration for the BFO 280 cSt 1/19 sample is 45,064 ppm (4.5%), almost 10 times higher. Therefore, when blended, the resulting oil would more closely resemble the PAH composition of the BFO source as shown by the blended sample (NC-61-NOAA) at the bottom of the figure. Both bunker fuel oils have a similar weight percentage of total aliphatic hydrocarbons; however, the MFO is significantly skewed to the higher-molecular-weight (and more persistent) n-C<sub>19</sub> to n-C<sub>37</sub> range. The S/T composition of BFO 280 cSt 1/19 is devoid of T18-oleanane and the TSSt is very low at only 40  $\mu\text{g/g}$ . MFO 280 11/28 contains T18-oleanane and a full suite of other steranes and triterpanes, and the TSSt is significantly higher at 1,872  $\mu\text{g/g}$ . Thus, a blend of these oils would present an SHC pattern that would reflect contributions from both sources, while the S/T pattern would more closely resemble MFO 280 11/28. These characteristics are reflected in the patterns at the bottom of the figure for the blended oil (NC-61-NOAA) collected from the beach adjacent to the vessel on February 14, 1999.

Figure 2 illustrates that the PAH histogram of one of the two diesels (MDO 1/19) is predominated by naphthalenes (N-N4), while the other diesel (MDO SK Corp 11/28) is predominated by alkylated phenanthrenes/anthracenes (P/A). Both diesels contain between 3.9-5.8% by weight PAH, so that would be the expected aromatic content in any diesel released from the vessel. While the SHC pattern for the MDO 1/19 sample is more characteristic of the narrower distillate range (n-C<sub>10</sub> to n-C<sub>25</sub>) normally associated with diesel oils, the n-alkane distribution for MDO SK Corp looks more like that of a fresh crude oil (n-C<sub>8</sub> through n-C<sub>32</sub>), although the TSHC is nearly 2.5 times lower at only 62,500 ppm (6.2%). MDO 1/19 contains almost no S/T with a TSSt of 31 ppm, while MDO SK Corp contains a full suite of S/T at a TSSt of 746 ppm. The PAH distribution for the Engine Room oil collected in July 1999 shows predominant naphthalenes (N-N4) and phenanthrenes/anthracenes (P-P/A3), a full suite of n-alkanes from n-C<sub>10</sub> through n-C<sub>34</sub>, and significant concentrations of S/T (at 1,618 ppm) with T15-norhopane and T19-hopane predominating over T18-oleanane.

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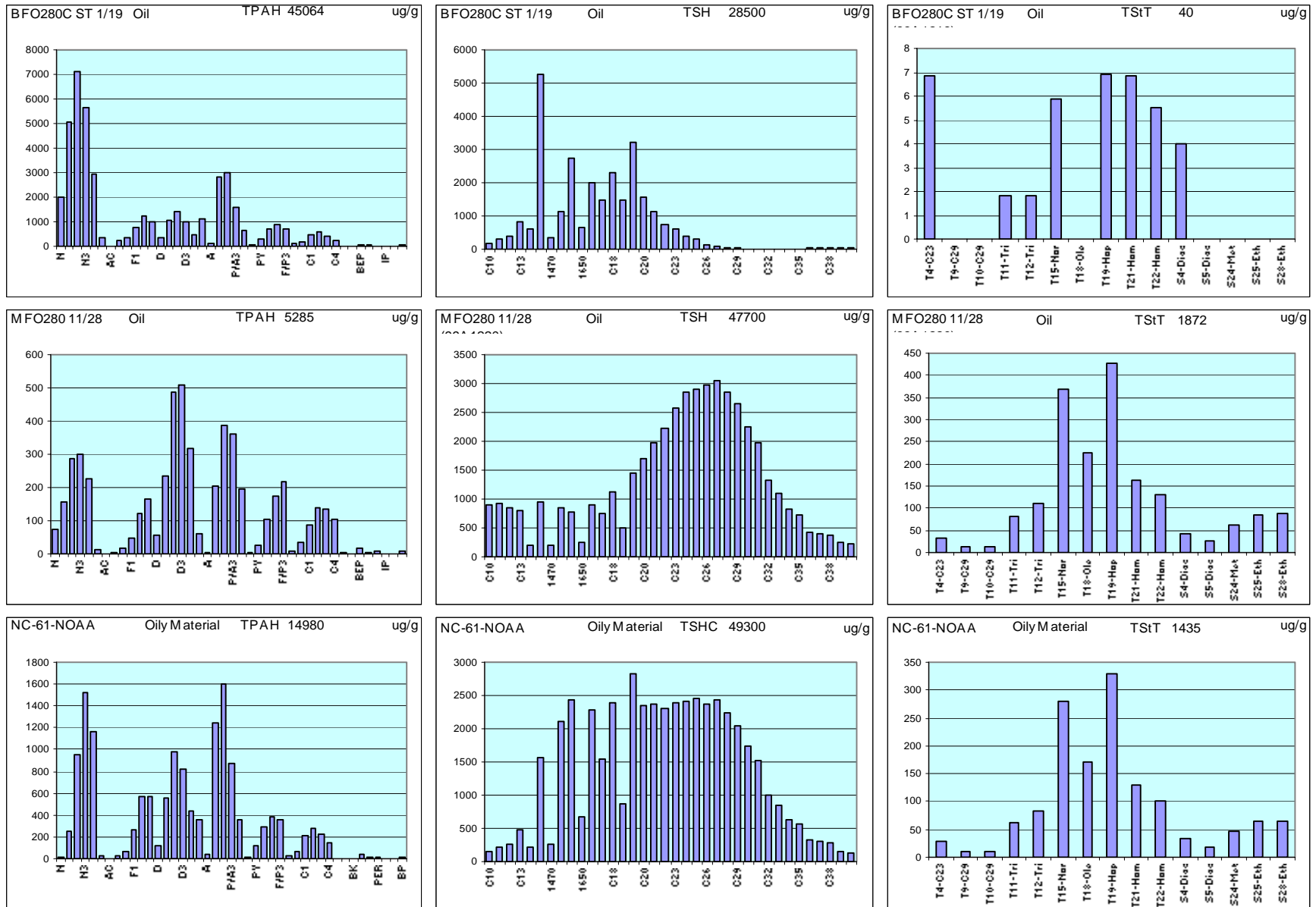


Figure 1. PAH, SHC, and S/T histogram plots of *New Carissa* Bunker fuels (BFO 280 CST 1/19 and MFO 280 11/28) and beached composite blend of oil (NC-61-NOAA) collected adjacent to the vessel at the time of the spill on Feb. 14, 1999. All concentrations are  $\mu\text{g/g}$  (ppm), and abbreviations for target analytes are identified in Table 1.

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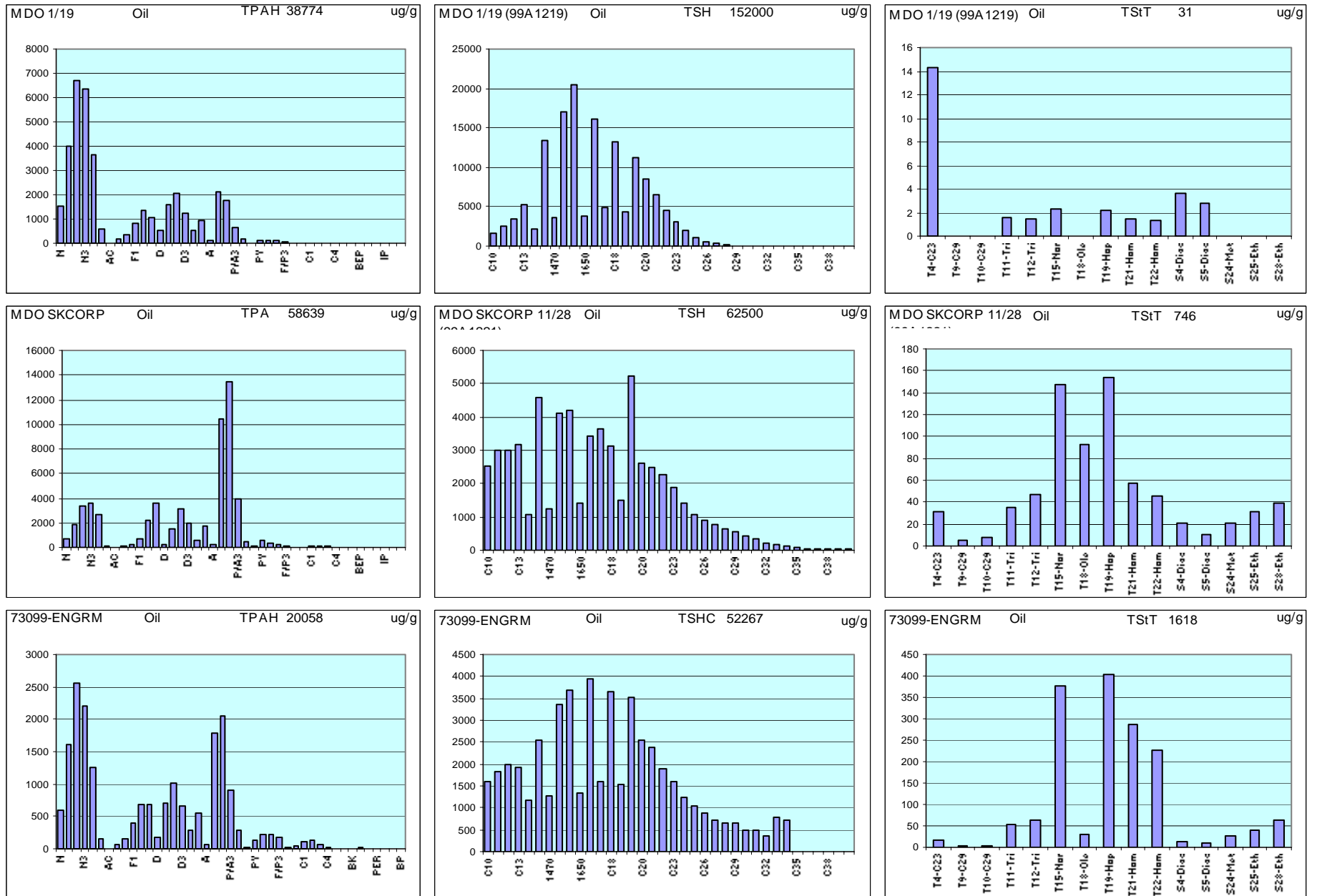


Figure 2. PAH, SHC, and S/T histogram plots of *New Carissa* diesel fuels (MDO 1/19 and MDO SK Corp) and engine oil collected from the stern of the vessel on 30 July 1999. All concentrations are µg/g. Abbreviations for target analytes are identified in Table 1.

With these drastically different source-oil characteristics, several analytic approaches were required for source identifications on the collected feather samples. The classic double-ratio plot of dibenzothiophenes to phenanthrenes/anthracenes (C2DBT/C2PA versus C3DBT/C3PA) was not helpful (Figure 3); the ratios were too similar between the primary sources (MFO, BFO, and MDOs) to provide any useful discrimination. However the alternative log-log plot of dibenzothiophenes to chrysenes (log C2DBT/C2Chr versus log C3DBT/C3Chr) was instrumental in confirming the uniqueness of the primary sources and blended nature of others (Figure 4).

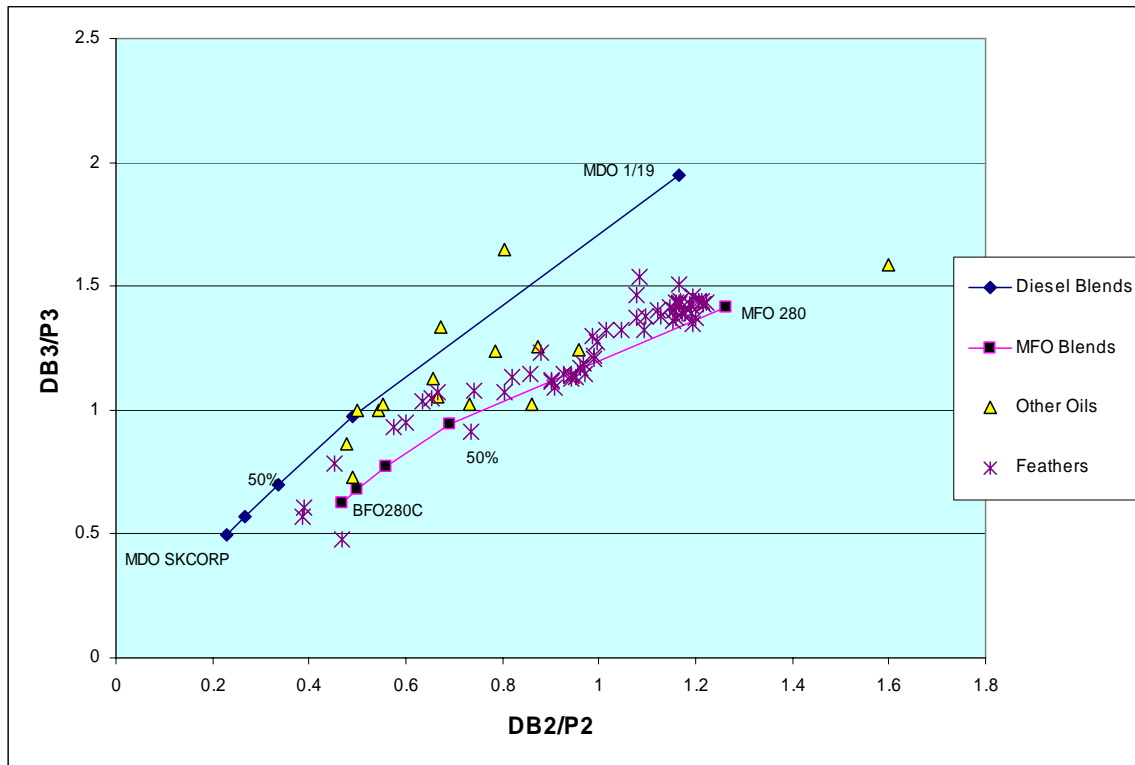


Figure 3. Double ratio plot of C2DBT/C2PA versus C3DBT/C3PA.

When the other oil and feather samples were overlaid on the double-ratio plot (Figure 5), it became apparent that there were 3 primary groups of oiled feathers: 1) feathers containing known source oils from the *New Carissa* (plotting on or near the BFO-MFO blend line); 2) feathers with oil from an unknown but similar source (lower left cluster); and 3) feathers with totally dissimilar oil(s) (lower left extreme outliers).



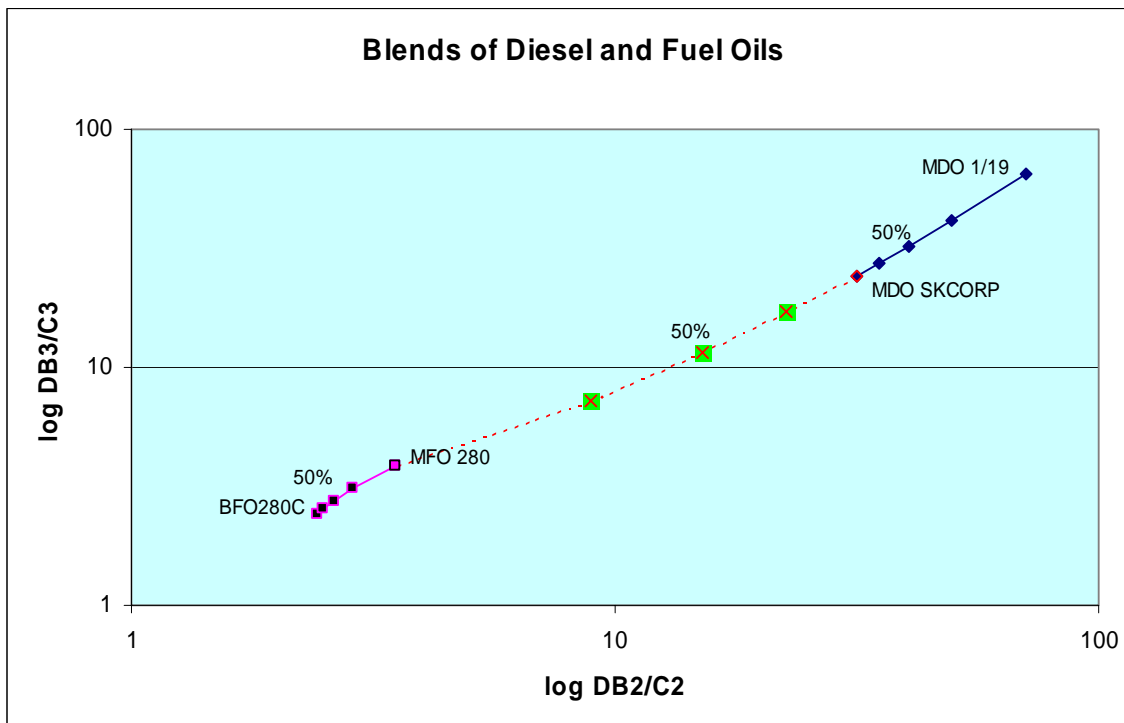


Figure 4. Double-ratio plot of log DBT3/C3 vs. log DBT2/C2 showing characteristics of blending source oils.

Using the blend optimization routines, it was possible to reduce the 19 source oils to just 5 primary types combined in various mixtures and states of weathering (Table 2). As one might expect from the vessel's tank capacities, the MFO and BFO oils were the dominant sources. Figures 6 and 7 present examples of the results from the blend optimization program used to generate the source blend values presented in Table 2. Figure 6 compares the PAH, SHC, and S/T patterns obtained where mixtures of MFO 280 and BFO 280 were combined to match the pattern displayed by the NC-61-NOAA sample collected along the beach adjacent to the bow of the *M/V New Carissa* immediately after it broke up in February 1999. As illustrated by the overall fit of all three analyte groups, allowing for evaporative weathering and dissolution of naphthalene (N) the 68:32 blend of MFO 280 and BFO 280 yields a pattern that is nearly identical to that obtained in the field sample. Figure 7 shows the results of blending the MDO SK Corp and MDO 1/19 samples to match the oil sample collected from the Stern Fuel Tank No. 5 on May 18, 1999. In this case, the 27:73 blend of the MDO SK Corp and MDO 1/19 diesel sources yields an excellent match to the patterns obtained on the field sample collected from the vessel.

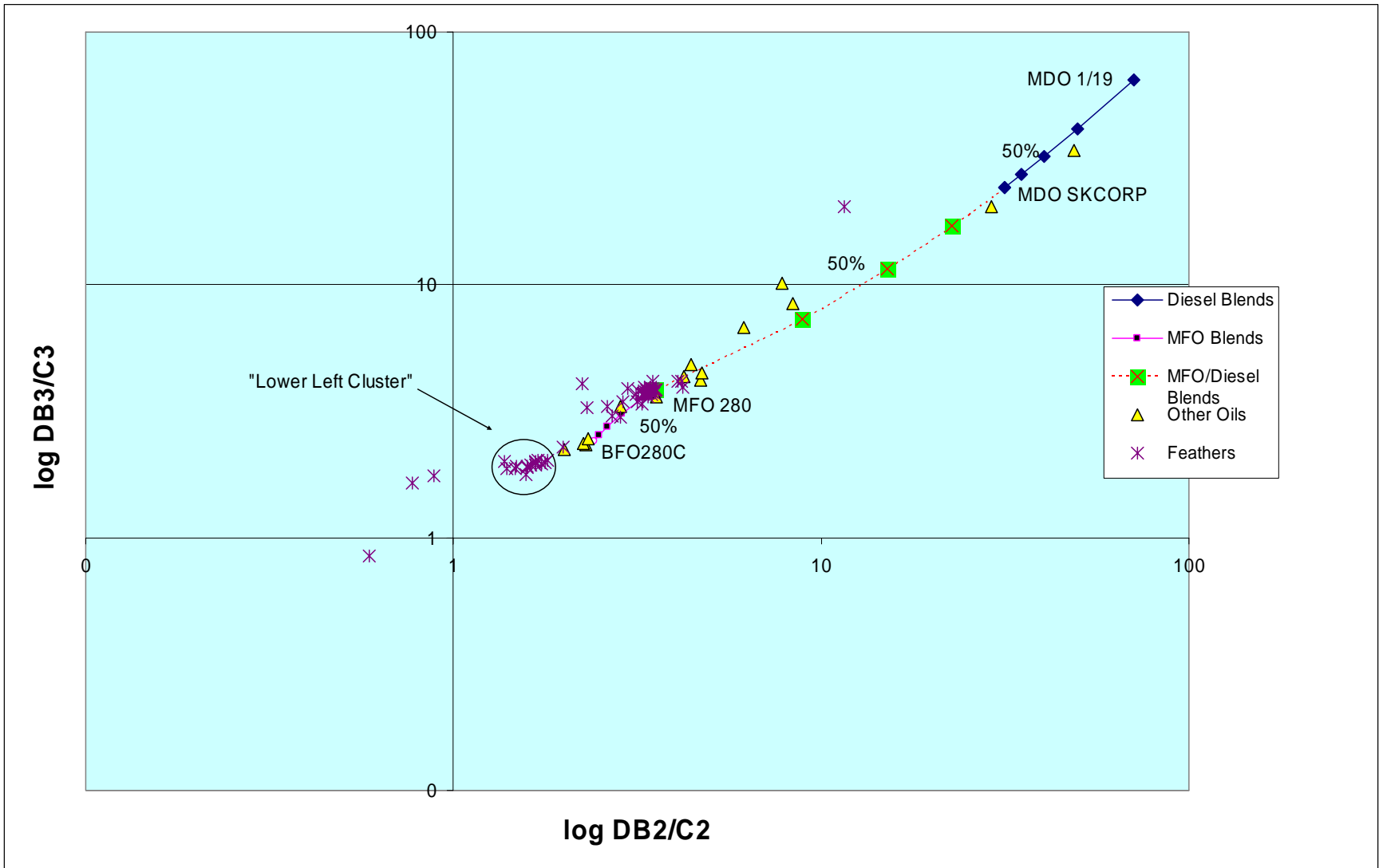


Figure 5. Double ratio plot of  $\log \text{DBT3/C3}$  vs.  $\log \text{DBT2/C2}$  including feather and source oil samples.

**Table 2. Characterization of *New Carissa* Oils as Blends from Five Primary Sources**

	Concentration (µg/g)			Source Blend					Comments
	Total PAH	Total St/T	Total SHC	MFO 280	BFO 280	MDO SK Corp	MDO 1/19	Engine Room	
<b>Source Oils</b>									
BFO280C ST 1/19	45064	40	28500		1.00				Archived original fuel sample.
MDO 1/19	38774	31	152000				1.00		Archived original fuel sample.
MFO280 11/28	5285	1872	47700	1.00					Archived original fuel sample.
MDO SKCORP 11/28	58639	746	62500			1.00			Archived original fuel sample.
73099-ENGRM	20058	1618	52267					1.00	Engine Room Oil
<b>Blends or Duplicates</b>									
99-130-04 BFO	58921	54	24506		1.00				ADL analysis of USCG split. 100% Battelle match
99-130-06 MDO 1/19	52494	42	140090				1.00		ADL analysis of USCG split. 100% Battelle match
99-130-05 MFO 280	5327	1372	41400	0.98	0.02				ADL analysis of USCG split. 98% match to Battelle data.
99-130-07 MDO SK	54554	681	88566			0.86	0.14		ADL analysis of USCG split. 86% match to Battelle data
RH-5F0-51899	48665	426	63493			0.27	0.73		Stern Fuel Tank #5. Blend of MDO SK Corp and MDO 1/19
Hold 2	38250	209	18759		1.00				S/T has correct fingerprint, but higher concentrations
Tank1/Hold2	10853	871	28610	0.85	0.15				Blend of MFO 280 & BFO 280
Tank2/Hold3	40560	34	17889	0.01	0.99				Almost pure BFO 280 (possible trace of MFO 280)
Fuel Tank #2	41018	37	17728	0.01	0.99				Almost pure BFO 280 (possible trace of MFO 280)
ST-1	12258	2085	14630					1.00	Engine Room Oil
NC-001	11383	2042	14119					1.00	Identical match to sample ST-1 (Engine Room Oil)
ET-1	5715	1245	33640	0.99	0.01				Blend of MFO 280 & BFO 280
ET-2	7739	1791	44900	0.94	0.06				Blend of MFO 280 & BFO 280
NC-61-NOAA	14980	1435	49300	0.68	0.32				Blend of MFO 280 & BFO 280
North Slope Crude	12038	621	14980						Potential non- <i>New Carissa</i> source

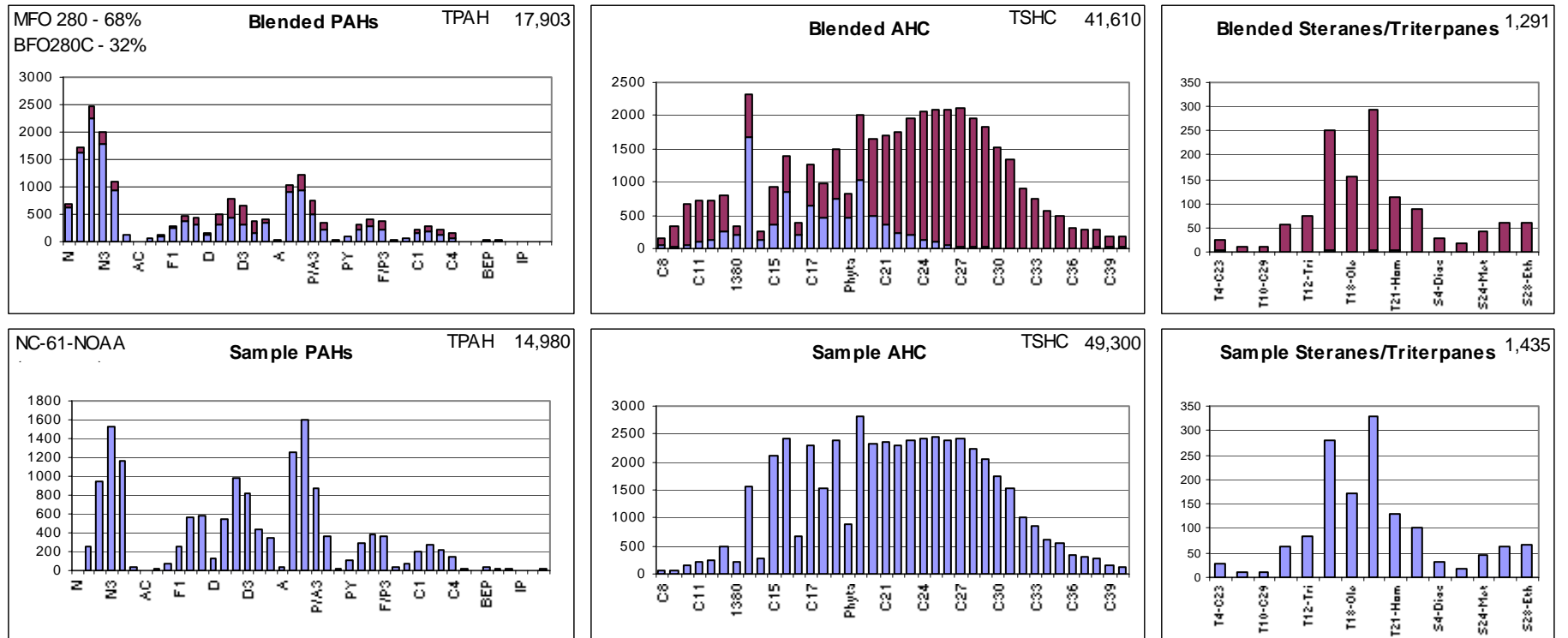


Figure 6. Results of the Blend Program combining MFO 280 and BFO 280 (68:32) to yield the histograms on the top (the MFO 280 contribution is denoted by the red and the BFO 280 by the blue) compared to the histograms obtained on the field sample NC-61-NOAA collected adjacent to the vessel on 14 February 1999. All concentrations are  $\mu\text{g/g}$ .

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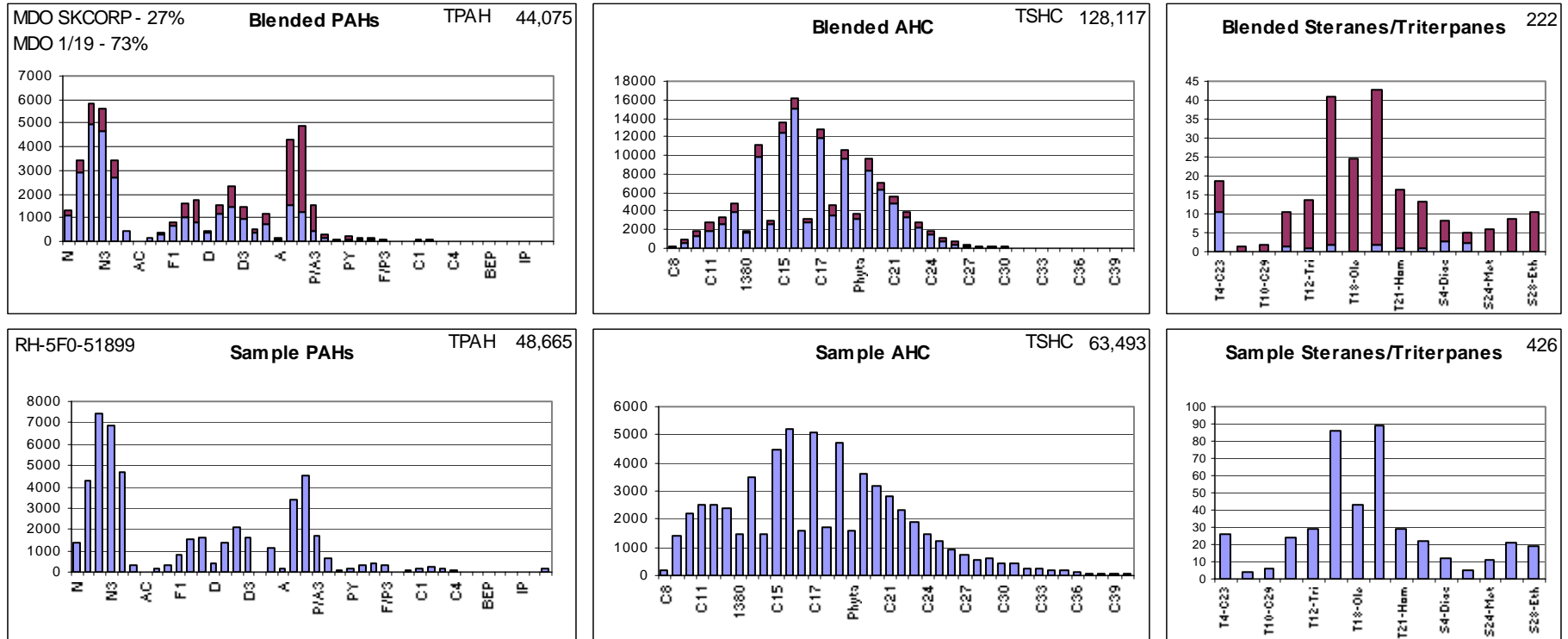


Figure 7. Results of the Blend Program combining MDO SKCORP and MDO 1/19 (27:73) to yield the histograms on the top (the MDO SKCORP contribution is denoted by the red and the MDO 1/19 by the blue) compared to the histograms obtained on the field sample RH-5F0-51899 collected from the Stern Fuel Tank No 5 on May 18, 1999. All concentrations are µg/g.

Stability of Reference Oils and Comparisons with Earlier Analyses

In undertaking these feather analyses, the Trustees were also concerned about 1) the archival stability of the reference oils and feather samples (both of which have been stored frozen for almost three years) and 2) the possible introduction of systematic errors by the fact that the initial analyses were completed by A.D. Little (ADL) while the most recent analyses were completed by Battelle New England Laboratories. Regarding the latter, both laboratories fortunately utilized analytical protocols based on NOAA Status and Trends procedures (Lauenstein and Cantillo 1993), and as such, they were essentially identical.

With regard to reference oil and sample integrity, histogram profiles and reported concentrations of testament oil samples run by both laboratories were compared for all three analyte groups (PAH, SHC, and S/T). Over the last three years, the histogram profiles have remained essentially proportionally identical, thus validating the qualitative comparisons over time and between both laboratories. For example, the blended data fits for the four primary reference oils (Table 2) illustrate a near perfect fit (better than 98%) between the two labs for three of the four reference oils. For the one deviant sample (MDO SKCorp), the ADL analysis gave an 86% fit with the Battelle data based on the relative comparison of individual analytes (see below). Although these summary fit data are impressive, the proportional differences between the laboratories’ results for individual analytes was the measure most relevant to the oil identification task; it was no greater than  $\pm 3\%$  for any given analyte.

Furthermore, as shown by the concentration data for the four reference oil samples (Table 3), the quantitative data are also directly comparable. Some quantitative differences do appear in Total PAH and Total SHC in Table 3 (for MDO 1/19 PAHs and MDO SKCORP SHCs), but these may be related to surrogate recovery-correction issues. Fortunately, neither factor had any direct impact on either the analyses or the interpretation of the results. The overall relative ratios of PAH:SHC:S/T for each reference oil are also very consistent. In summary, based on the data analysis of the feather samples considered in this report, we can confidently state that when there was a positive match for known *New Carissa* source oils, there was no evidence of sample degradation (additional evaporative losses or microbial degradation) during frozen storage.

**Table 3. Comparison of Reference Oil Analyses at A.D. Little and Battelle New England Laboratories (all concentrations in  $\mu\text{g/g}$ )**

Reference Oils	Total PAH		Total SHC		Total S/T	
	Battelle	ADL	Battelle	ADL	Battelle	ADL
BFO280C ST 1/19	45,064	55,953	28,500	25,491	40	32
MDO 1/19	38,774	51,719	152,000	152,242	31	22
MFO280 11/28	5,285	5,402	47,700	47,470	1,872	1,414
MDO SKCORP 11/28	58,039	58,279	62,500	102,640	746	603

Feather Matches

Forty-one samples were positively identified as matching known *New Carissa* source oils, and one was identified as a probable match. These are listed below (Table 4). Complete details for each sample are presented in Appendix A, and Appendix B presents histogram plots for all of the analyzed feathers and

known *New Carissa* source oils considered in this analysis. Only the heavier bunker oils (MFO 280 and BFO 280) were found on these feathers; there was no evidence of diesel oils. We believe that the heavy surf action at the time of the spill dispersed the lower viscosity diesel fuels into the water column, and as such, the diesel oils did not persist as a coherent surface slick that would coat or otherwise contaminate birds at, or diving through, the air-sea interface. Figures 8 and 9 present characteristic examples of the PAH, SHC, and S/T histogram plots typical of the feathers that were positively identified as being contaminated with *New Carissa* oil. Figure 8 shows the pattern obtained where primarily one fuel was responsible for the contamination, and Figure 9 shows the pattern for a feather that was clearly contaminated by a blend of the two fuels.

**Table 4. Feather samples positively identified as being contaminated with M/V *New Carissa* oil**

01MS03	16MS03	29MS03	45MS03	57MS03	69MS03
02MS03	17MS03	30MS03	46MS03	58MS03	54MS03*.
06MS03	18MS03	32MS03	48MS03	59MS03	
08MS03	20MS03	34MS03	49MS03	60MS03	
09MS03	21MS03	35MS03	50MS03	61MS03	
11MS03	22MS03	36MS03	52MS03	63MS03	
13MS03	25MS03	41MS03	55MS03	65MS03	
15MS03	28MS03	42MS03	56MS03	67MS03	

\*Note that sample 54MS03 was assessed as a probable *New Carissa* sample (see Appendix A).

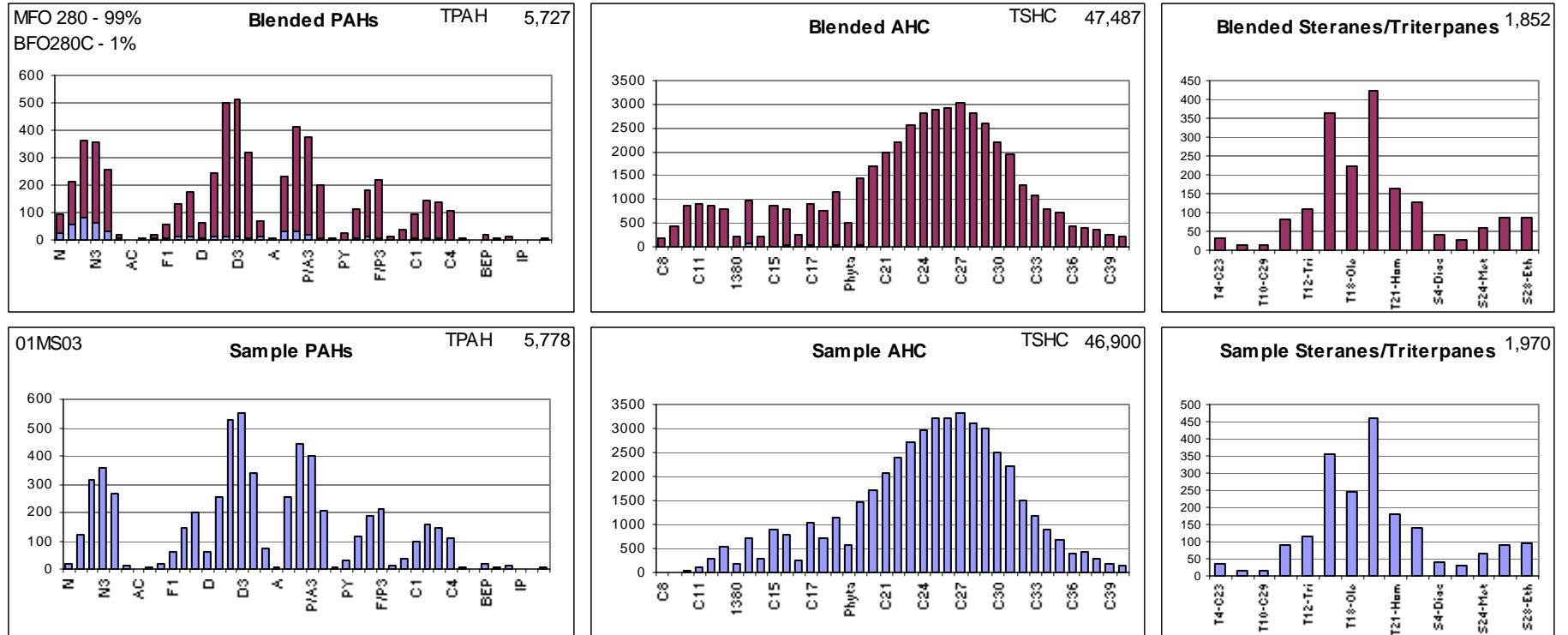


Figure 8. Example of the PAH, SHC, and S/T histogram matches between sample 01MS03, which was characterized as being contaminated with essentially pure MFO 280 with a blend of 99% MFO 280 and 1 % BFO 280. This sample is characteristic of most of the samples classified as contaminated with *M/V New Carissa* oil in Table 4 and Appendix A when the MFO 280 content was 95% or higher. The loss of naphthalene (N) and n-alkanes below n-C<sub>14</sub> in the sample can be easily attributed to natural weathering processes (Payne and McNabb, Jr. 1984; Payne et al. 1984). All concentrations are µg/g.



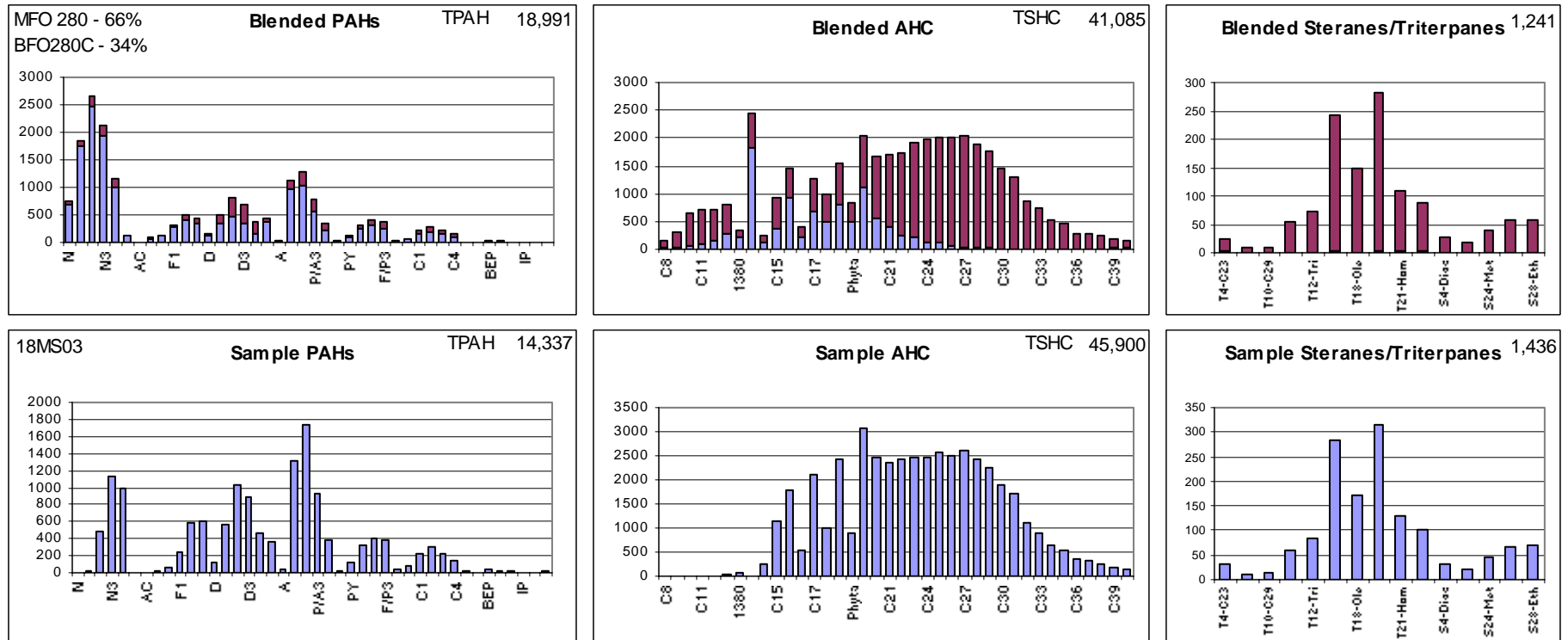


Figure 9. Example of a match between an oiled feather (sample 18MS03) and a blend of 66% MFO 280 and 34% BFO 280. Allowing for evaporation and dissolution weathering losses of the naphthalenes (N – N4) and evaporation weathering of n-alkanes below n-C<sub>15</sub>, a very close match is obtained that is characteristic of many of the blended matches presented in Appendix A. All concentrations are µg/g.

Feather Outliers

Table 5 lists the nineteen samples that could not be matched with the known oils from the *New Carissa* spill. The first eleven samples were from a single unknown source; the PAH, SHC, and S/T histogram patterns at the top of Figure 10 are representative of those samples. These samples clustered in the lower left corner of the double-ratio plot shown in Figure 5, contained too much F/P and C, and didn't have enough T18-oleanane to match any of the known *New Carissa* oils or blends.

Three other samples appeared similar to the previous nineteen in many respects but slightly different; the middle and bottom set of histograms in Figure 10 illustrate the subtle differences observed in those samples. Specifically, sample 07MS03 has a similar PAH and S/T pattern, but the SHC group shows a bimodal distribution of higher molecular weight (n-C<sub>22</sub> to n-C<sub>29</sub>) n-alkanes. Sample 23MS03 (on the bottom in Figure 10) has a similar SHC and S/T pattern compared to sample 03MS03, but the PAH pattern is not the same, with more dibenzothiophenes than phenanthrenes, and much higher fluoranthenes/pyrenes (F/P1-F/P3) and chrysenes (C1-C4).

The remaining five samples in Table 5 represented five entirely unique sources not matching any of the provided reference oils. Figure 11 presents the PAH, SHC, and S/T histogram plots for samples 14MS03, 31MS03, and 66MS03. In the double-ratio plot shown in Figure 5, samples 31MS03 and 66MS03 are outliers separated to the far left from all other samples while sample 14MS03 plots by itself in the upper middle portion of the figure above the "MFO/Diesel Blends" line. Note that while the remaining two unique samples, 12MS03 and 70MS03 do not appear as outliers in the double-ratio plot, their histogram signatures confirm their uniqueness (see Appendix A).

**Table 5. Contaminated feather samples that were not consistent with the available M/V *New Carissa* source oils or blends**

Unknown Source 1		Similar to Source 1	Unique Sources
03MS03	38MS03	07MS03	12MS03
05MS03	43MS03	23MS03	14MS03
10MS03	47MS03	51MS03	31MS03
19MS03	62MS03		66MS03
33MS03	68MS03		70MS03
37MS03			

The S/T histograms for all three samples shown in Figure 11 do not contain sufficient T-18 oleanane relative to the other S/T to be consistent with known *New Carissa* source oils or blends, and the PAH and SHC patterns are each unique and unlike any of the available sources. Sample 14MS03 contains higher concentrations of dibenzothiophenes relative to chrysenes which is inconsistent with known *New Carissa* oils, and the SHC pattern shows extensive biodegradation of the n-alkanes relative to the isoprenoids, pristane and phytane. Sample 31MS03 contains too much chrysene in addition to the incorrect S/T pattern compared to known *New Carissa* oils and blends, and sample 66MS03 is characterized by histogram plots that taken together aren't consistent with any of the available *New Carissa* sources or any of the other feather samples examined.

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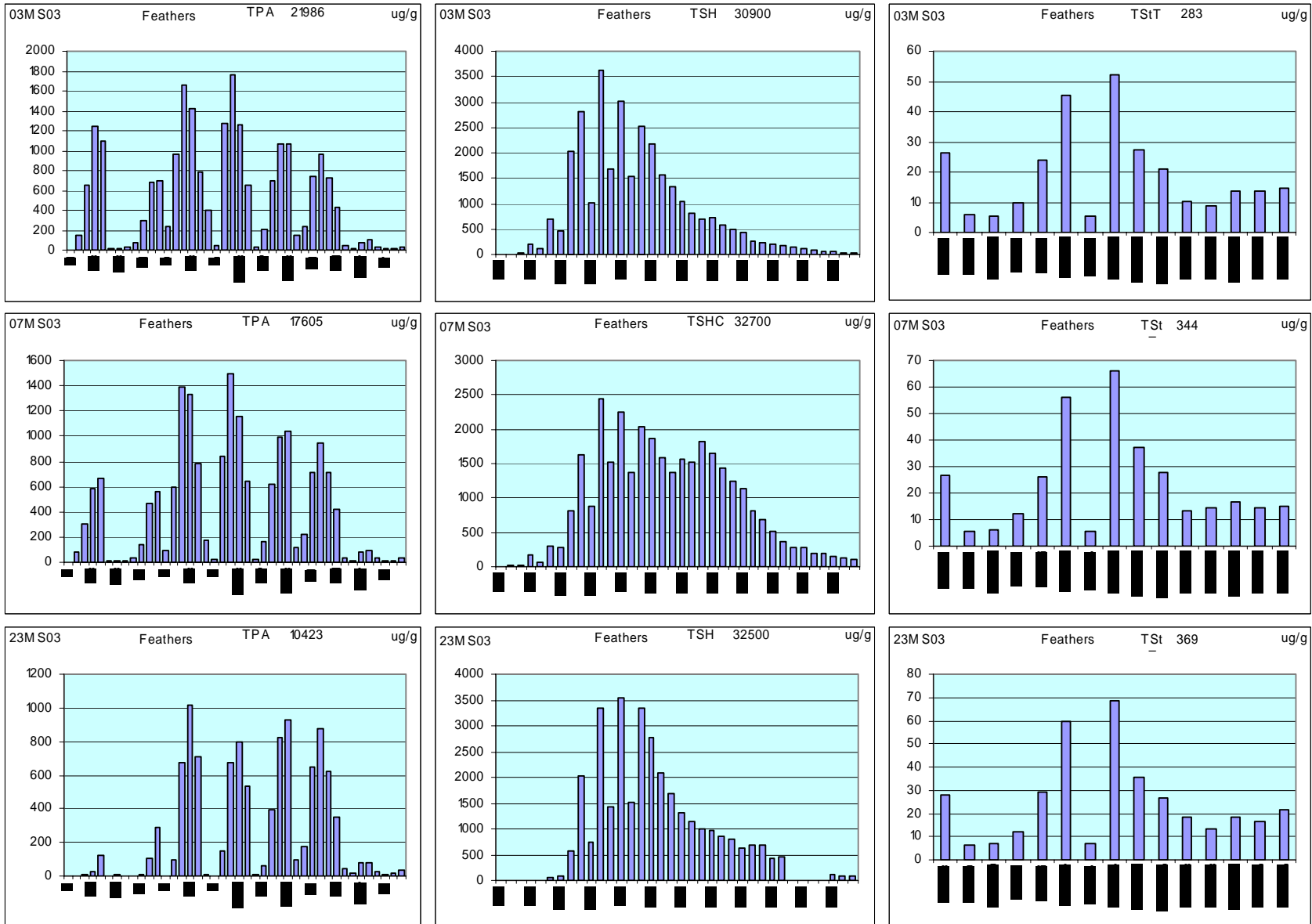


Figure 10. Representative histograms of eleven common samples (top row, sample 03MS03) that were not consistent with known *New Carissa* oils or blends, and three other samples that were similar to but not a match with 03MS03 (see text, Table 5, and Appendix A).

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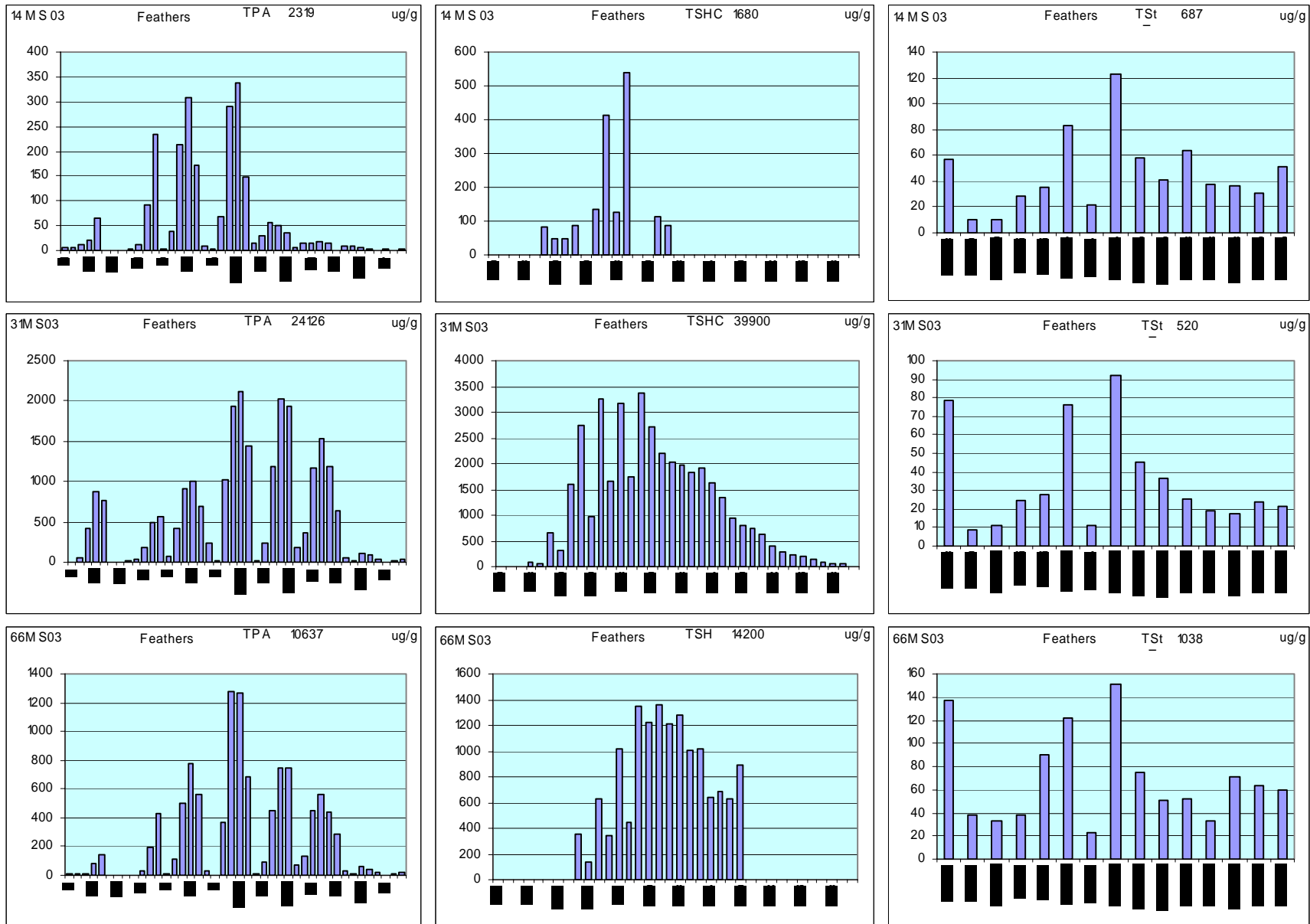


Figure 11. Representative histograms of feathers contaminated with unique oil sources that were not consistent with known *New Carissa* oils or blends, and that did not match or group with each other or any of the other samples examined (see text, Table 5, and Appendix A).

## Conclusions

Based on comparisons of analyte histograms and double-ratio plots with the known *New Carissa* reference oils, forty-one of the sixty-one feathers sampled had oil conclusively identified from the *New Carissa* spill. One additional sample was considered a probable match to the *New Carissa* reference sources. The remaining nineteen feather samples were not consistent with any of the provided reference oils. They appeared to represent an additional 6 or 7 non-referenced oil sources.

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Appendix A Client Sample ID	New Carissa Source	Concentration (µg/g)			Double Ratio Plot Outlier	Source Blend					Comments
		Total PAH	Total St/T	Total SHC		MFO 280	BFO 280	MDO SK Corp	MDO 1/19	Engine Room	
<b>Reference Oil Samples</b>											
BFO280C ST 1/19	yes	45064	40	28500			1.00				Pure source oil used to blend and match other samples
MDO 1/19	yes	38774	31	152000				1.00			Pure source oil used to blend and match other samples
MFO280 11/28	yes	5285	1872	47700		1.00					Pure source oil used to blend and match other samples
MDO SKCORP 11/28	yes	58639	746	62500				1.00			Pure source oil used to blend and match other samples
ET2	yes	7739	1791	44900		0.94	0.06				Blend of MFO 280 & BFO 280 (94:6)
NC-61-NOAA	yes	14980	1435	49300		0.68	0.32				Blend of MFO 280 & BFO 280 (68:32)
North Slope Crude	no	12038	621	14980							Separate oil fingerprint used to rule out other sources (passing tankers, etc.)
99-130-04 BFO	yes	58921	54	24506			1.00				ADL analysis of USCG split. 100% Battelle match
99-130-06 MDO 1/19	yes	52494	42	140090				1.00			ADL analysis of USCG split. 100% Battelle match
99-130-07 MDO SK	yes	54554	681	88566				0.86	0.12		ADL analysis of USCG split. 86% match to Battelle data
99-130-05 MFO 280	yes	5327	1372	41400		0.98	0.02				ADL analysis of USCG split. 98% match to Battelle data.
ET-1	yes	5715	1245	33640		0.99	0.01				Blend of MFO 280 & BFO 280 (99:1)
NC-001	yes	11383	2042	14119						1.00	Identical match to sample ST-1 (Engine Room Oil)
RH-5F0-51899	yes	48665	426	63493				0.27	0.73		Stern Fuel Tank #5. Blend of MDO SK Corp and MDO 1/19 (33:67)
ST-1	yes	12258	2085	14630						1.00	Engine Room Oil
HOLD 2	yes	38250	209	18759			1.00				Excellent fit for PAH and SHC, S/T has correct fingerprint, but higher concentrations
TANK1/HOLD2	yes	10853	871	28610		0.85	0.15				Blend of MFO 280 & BFO 280 (85:15)
TANK2/HOLD3	yes	40560	34	17889		0.01	0.99				Almost pure BFO 280 (possibly



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Appendix A Client Sample ID	New Carissa Source	Concentration (µg/g)			Double Ratio Plot Outlier	Source Blend					Comments
		Total PAH	Total St/T	Total SHC		MFO 280	BFO 280	MDO SK Corp	MDO 1/19	Engine Room	
											~ 1% MFO 280 also present)
FUEL TANK #2	yes	41018	37	17728		0.01	0.99				Almost pure BFO 280 (possibly ~ 1% MFO 280 also present)
73099-ENGRM	yes	20058	1618	52267					1.00		Engine Room Oil
<b>Feather Samples</b>											
01MS03	yes	5778	1970	46900		0.99	0.01				MFO 280
02MS03	yes	5308	1912	45300		0.99	0.01				MFO 280
03MS03	no	21986	283	30900	LLC						Not consistent with New Carissa oils. Unknown weathered crude or bunker oil. In lower left corner of DBT CHR plots.
05MS03	no	21356	298	33600	LLC						Not consistent with New Carissa oils. Unknown weathered crude or bunker oil. In lower left corner of DBT CHR plots.
06MS03	yes	9299	1894	52900		0.92	0.08				Blend of MFO 280 & BFO 280 (92:8) same pattern as ET 2
07MS03	no	17605	344	32700	LLC						Not consistent with New Carissa oils. Unknown weathered crude or bunker oil. In lower left corner of DBT CHR plots.
08MS03	yes	5748	1948	48000		0.99	0.01				MFO 280
09MS03	yes	6455	1924	46000		0.96	0.04				Blend of MFO 280 and BFO 280 (96:4)
10MS03	no	19425	336	30200	LLC						Not consistent with New Carissa oils. Unknown weathered crude or bunker oil. In lower left corner of DBT CHR plots.
11MS03	yes	11918	1612	48800		0.77	0.23				Blend of MFO 280 & BFO 280 (77:23) same pattern as NC 61-NOAA
12MS03	no	37	64	12700							Unknown lube oil, not consistent with New Carissa oils. No DBT, P/A, or CHR.
13MS03	yes	5081	1953	47200		0.99	0.01				MFO 280
14MS03	no	2319	687	1680	Mid-Upper solo						Possibly very weathered ANS oil plus trace of something else. Heavy microbial degradation of

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Appendix A Client Sample ID	New Carissa Source	Concentration (µg/g)			Double Ratio Plot Outlier	Source Blend					Comments
		Total PAH	Total St/T	Total SHC		MFO 280	BFO 280	MDO SK Corp	MDO 1/19	Engine Room	
											SHC. Trace of T-18 oleanane.
15MS03	yes	4299	1567	44000		0.99	0.01				MFO 280
16MS03	yes	10695	1636	42700		0.85	0.15				Blend of MFO 280 & BFO 280 (85:15)
17MS03	yes	4274	1656	49600		0.99	0.01				Weathered MFO 280
18MS03	yes	14337	1436	45900		0.66	0.34				Blend of MFO 280 & BFO 280 (66:34) same pattern as weathered NC 61-NOAA
19MS03	no	24515	310	28700	LLC						Not consistent with New Carissa oils. Unknown weathered crude or bunker oil. In lower left corner of DBT CHR plots.
20MS03	yes	8331	2157	55300		0.95	0.05				Blend of MFO 280 & BFO 280 (95:5) same pattern as ET 2
21MS03	yes	536	232	5060		0.99	0.01				Weathered MFO 280
22MS03	yes	3078	504	14000		0.78	0.22				Blend of MFO 280 & BFO 280 (78:22) same pattern as weathered NC 61-NOAA
23MS03	no	10423	369	32500	LL solo						One of 3 outliers to left of 1 on DBT CHR plots. Way too much CHR and S/T pattern wrong.
25MS03	yes	4788	1724	48700		0.99	0.01				Weathered MFO 280
28MS03	yes	8808	1683	44100		0.9	0.1				Blend of MFO 280 & BFO 280 (90:10)
29MS03	yes	17932	1356	49500		0.63	0.37				Blend of MFO 280 & BFO 280 (63:37) same pattern as lightly weathered NC 61-NOAA
30MS03	yes	4241	2042	43100		0.98	0.02				Weathered MFO 280
31MS03	no	24126	520	39900	LL solo						One of 3 outliers to left of 1 on DBT CHR plots. Way too much CHR and S/T pattern wrong.
32MS03	yes	6374	1939	45400		0.99	0.01				MFO 280
33MS03	no	23791	310	30300	LLC						Not consistent with New Carissa oils. Unknown weathered crude or bunker oil. In lower left corner of DBT CHR plots.
34MS03	yes	4885	2208	59700		0.99	0.01				Weathered MFO 280

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Appendix A Client Sample ID	New Carissa Source	Concentration (µg/g)			Double Ratio Plot Outlier	Source Blend					Comments
		Total PAH	Total St/T	Total SHC		MFO 280	BFO 280	MDO SK Corp	MDO 1/19	Engine Room	
35MS03	yes	6134	2036	50000		1.00					Fresh MFO 280
36MS03	yes	4451	1813	48700		0.98	0.02				Weathered MFO 280
37MS03	no	25963	401	37700	LLC						Not consistent with New Carissa oils. Unknown weathered crude or bunker oil. In lower left corner of DBT CHR plots.
38MS03	no	25095	288	35200	LLC						Not consistent with New Carissa oils. Unknown weathered crude or bunker oil. In lower left corner of DBT CHR plots.
41MS03	yes	7954	1130	20700	LLC	0.7 est	0.3 est				Very weathered NC-61-NOAA pattern, but also in center of non-NC oils cluster in lower left corner of DBT CHR plots. ID verified in blend and grapher.
42MS03	yes	9505	2266	64300		0.68	0.32				Blend of MFO 280 & BFO 280 (68:32) same pattern as very weathered NC 61-NOAA
43MS03	no	25409	291	32300	LLC						Not consistent with New Carissa oils. Unknown weathered crude or bunker oil. In lower left corner of DBT CHR plots.
45MS03	yes	5098	3549	94100		0.97	0.03				Very weathered MFO 280
46MS03	yes	6114	1872	46600		0.99	0.01				Fresh MFO 280
47MS03	no	22674	400	33800	LLC						Not consistent with New Carissa oils. Unknown weathered crude or bunker oil. In lower left corner of DBT CHR plots.
48MS03	yes	5255	2202	52400		0.98	0.02				Very weathered MFO 280
49MS03	yes	4993	2398	59200		0.99	0.01				Very weathered MFO 280
50MS03	yes	6864	2072	55900		0.96	0.04				Weathered blend of MFO 280 and BFO 280 (96:4)
51MS03	no	22985	444	38300	LLC						Not consistent with New Carissa oils, but a little more T-18 oleane than other "nos." In lower left corner of DBT CHR plots.
52MS03	yes	6066	1926	45900		0.98	0.02				MFO 280

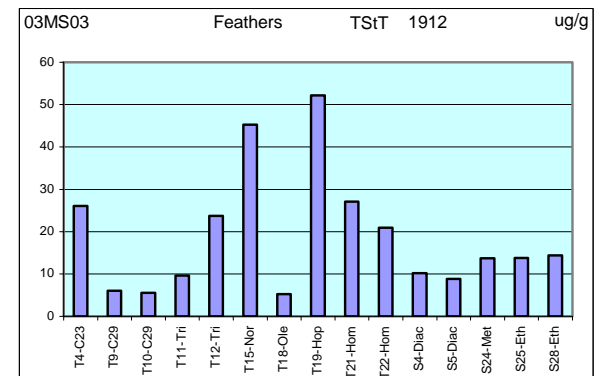
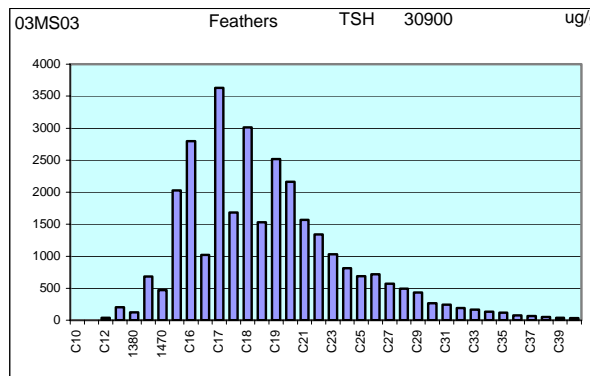
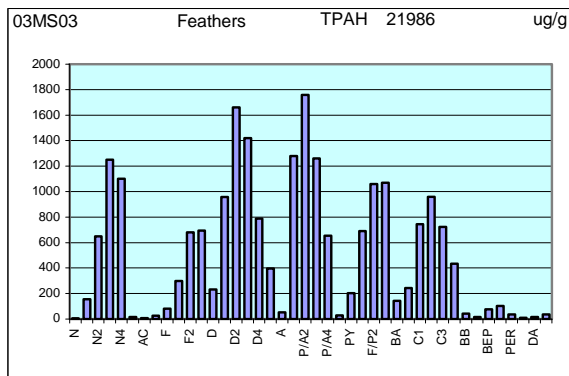
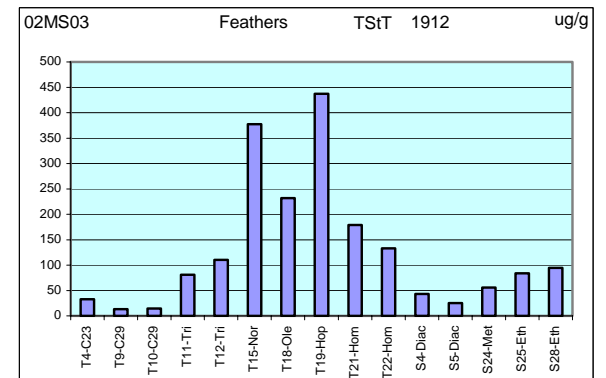
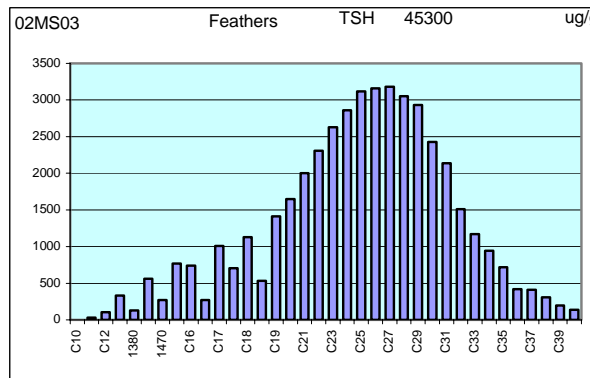
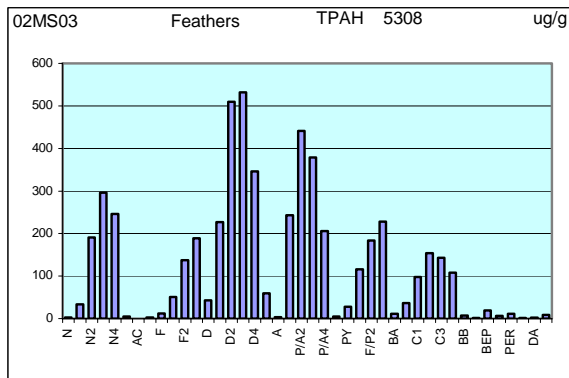
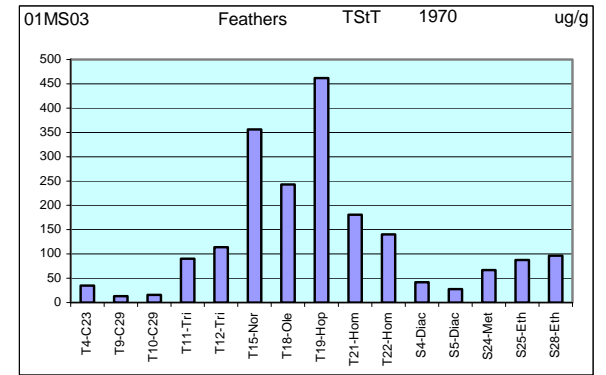
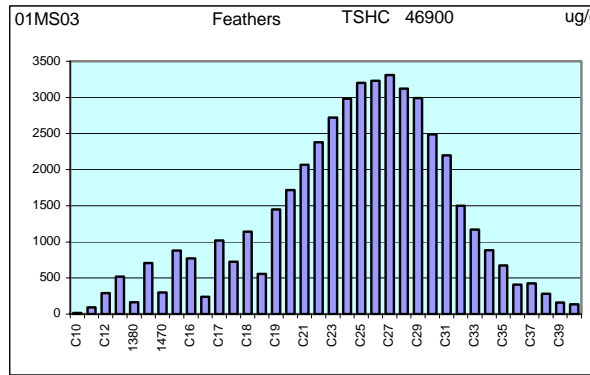
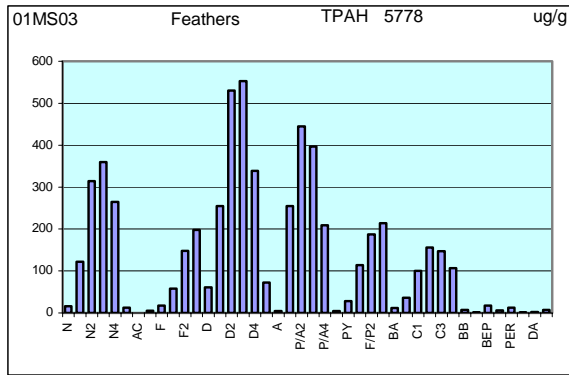
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Appendix A Client Sample ID	New Carissa Source	Concentration (µg/g)			Double Ratio Plot Outlier	Source Blend					Comments
		Total PAH	Total St/T	Total SHC		MFO 280	BFO 280	MDO SK Corp	MDO 1/19	Engine Room	
54MS03	probably	10467	900	42200	LLC	0.94	0.06				Yes, by blends. MFO 280 and BFO 280 (94:6). Near lower left corner of DBT CHR plots. Plots on top of Hold 2 source oil.
55MS03	yes	5909	2525	63900		0.99	0.01				Weathered MFO 280
56MS03	yes	11391	1746	48500		0.83	0.17				Lightly weathered blend of MFO 280 and BFO 280 (83:17).
57MS03	yes	4157	1772	45100		0.98	0.02				Weathered MFO 280
58MS03	yes	6522	1865	47400		0.87	0.13				Weathered ET-2 and MFO 280 blend (87:13).
59MS03	yes	6430	1959	50000		0.98	0.02				Weathered MFO 280
60MS03	yes	5850	2049	48400		0.98	0.02				Weathered MFO 280
61MS03	yes	5575	2030	53400		0.99	0.01				Weathered MFO 280
62MS03	no	25235	328	35500	LLC						Not consistent with New Carissa oils. Unknown weathered crude or bunker oil. In lower left corner of DBT CHR plots.
63MS03	yes	6260	2267	53300		1.00					MFO 280
65MS03	yes	5570	2255	51200		1.00					Weathered MFO 280
66MS03	no	10637	1038	14200	LL solo						One of 3 outliers to left of 1 on DBT CHR plots. Way too much CHR and S/T pattern wrong.
67MS03	yes	6479	2031	49900		0.99	0.01				Lightly weathered MFO 280
68MS03	no	23177	342	34700	LLC						Not consistent with New Carissa oils. Unknown weathered crude or bunker oil. In lower left corner of DBT CHR plots.
69MS03	yes	4606	2286	52600		0.94	0.06				Very weathered MFO 280
70MS03	no	12520	628	10200	LLC						Not consistent with New Carissa oils. Could be very very weathered BFO 280 plus 2% MFO 280, but S/T concentrations too high.

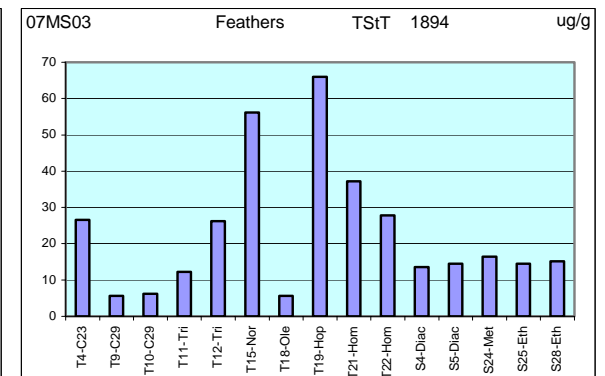
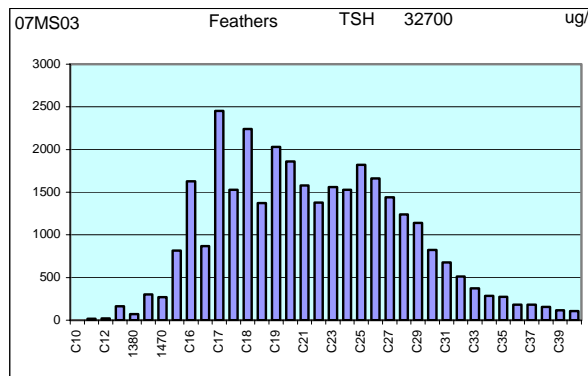
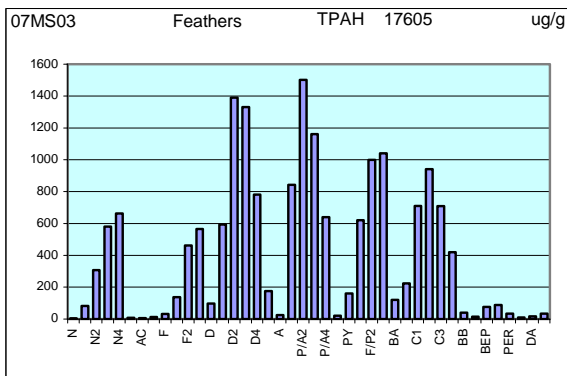
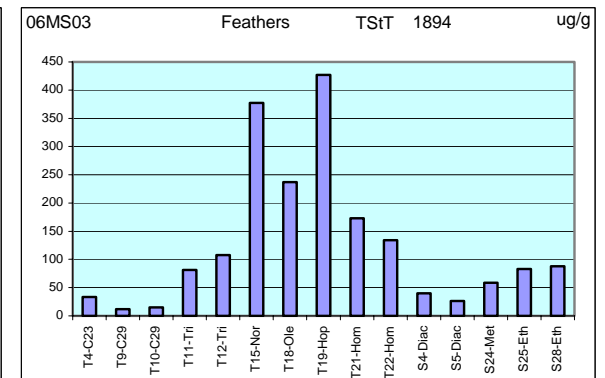
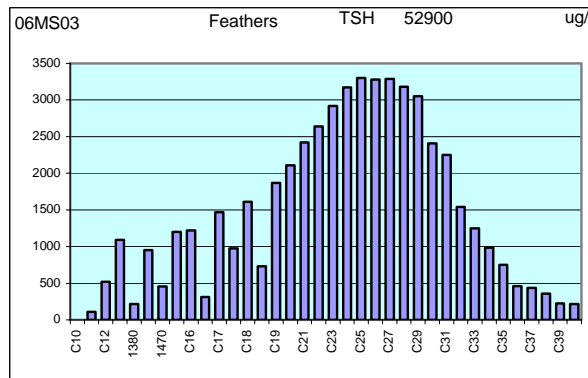
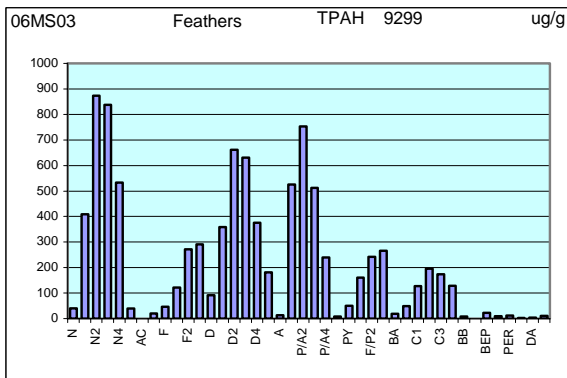
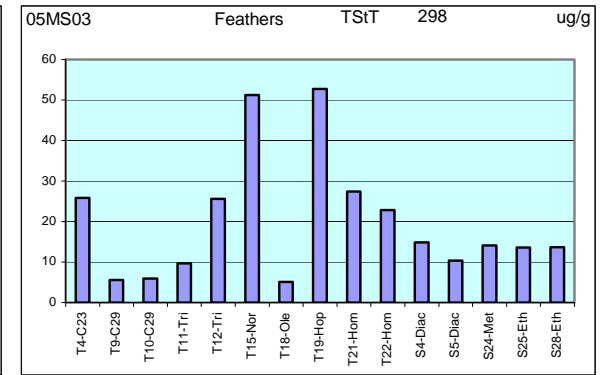
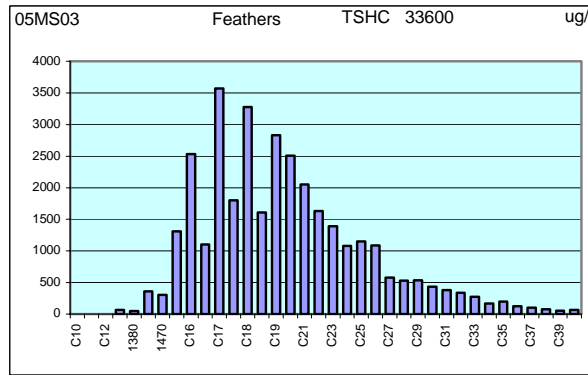
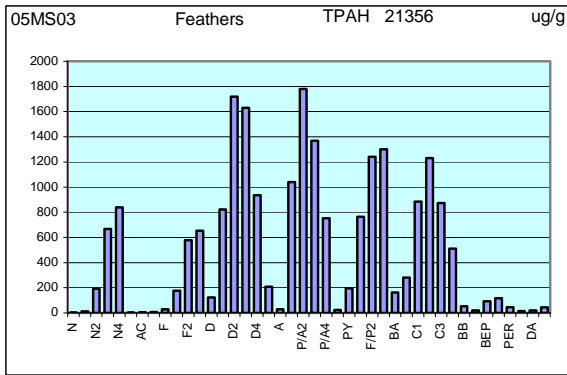
**Appendix B – Histogram plots of all feather samples and known *New Carissa* source oils.**

Each row of plots represents PAH, SHC and Sterane/Triterpane results for a single feather sample. Known *New Carissa* source oils and a North Slope Crude oil reference sample follow the feather samples.

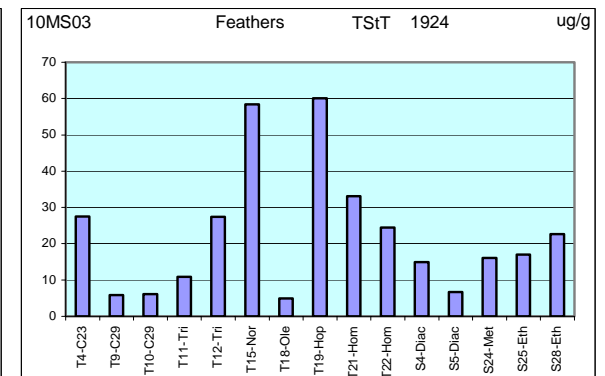
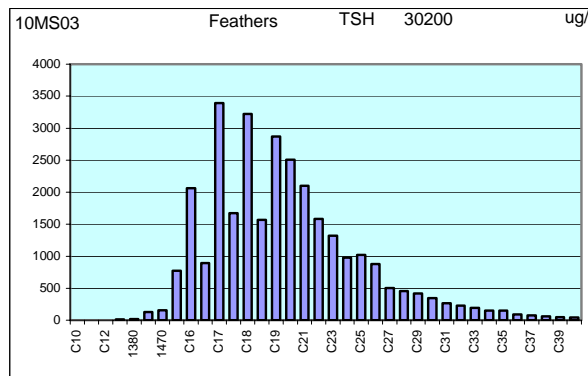
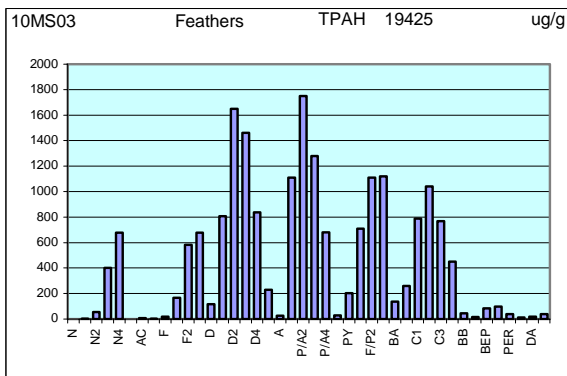
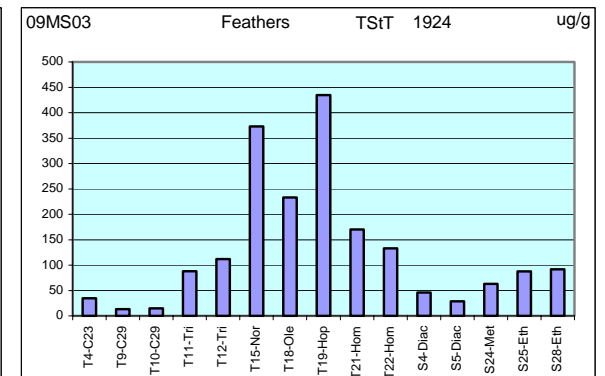
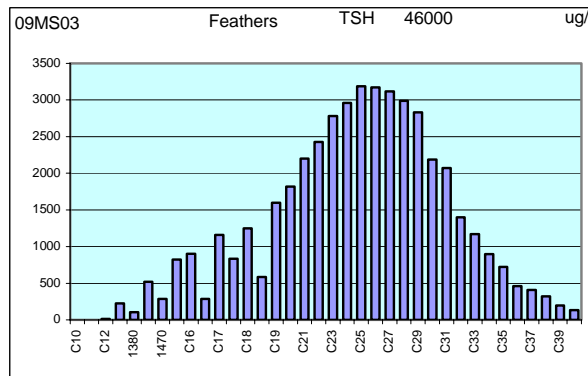
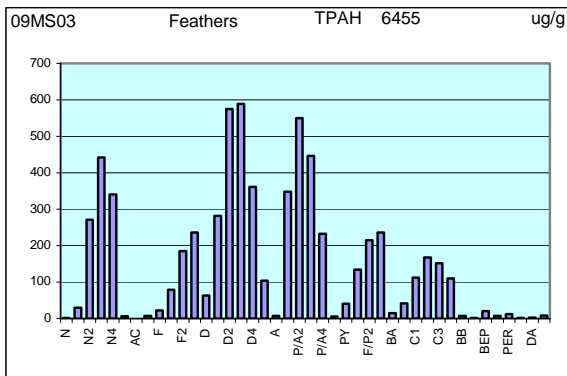
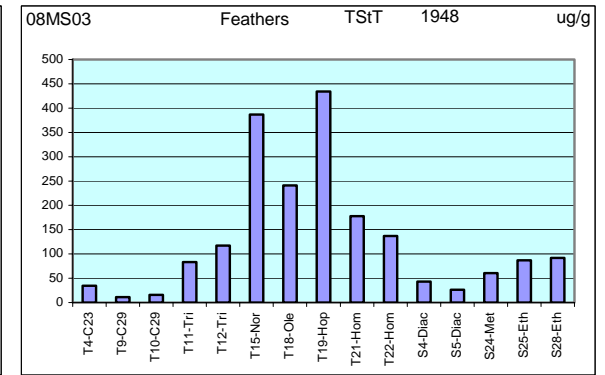
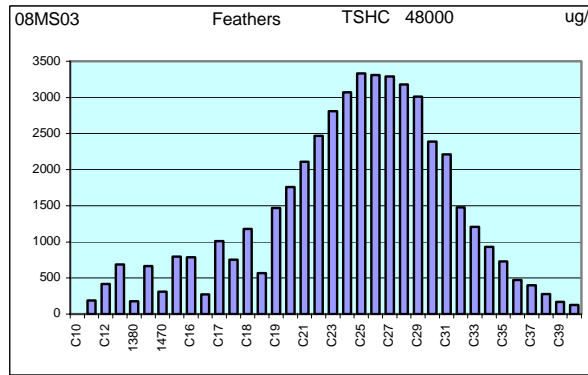
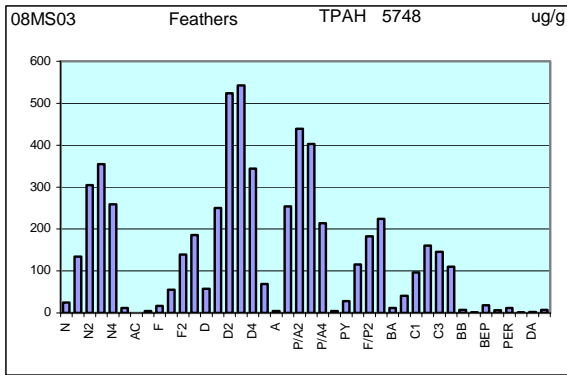
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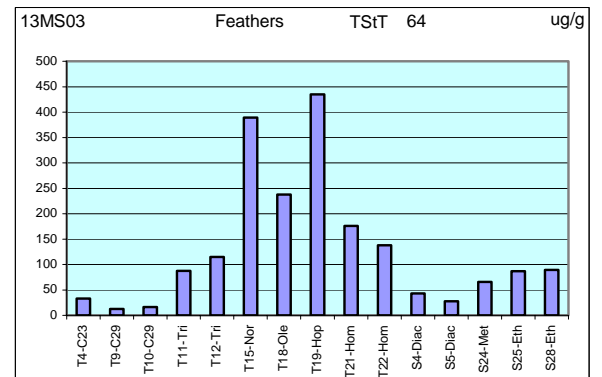
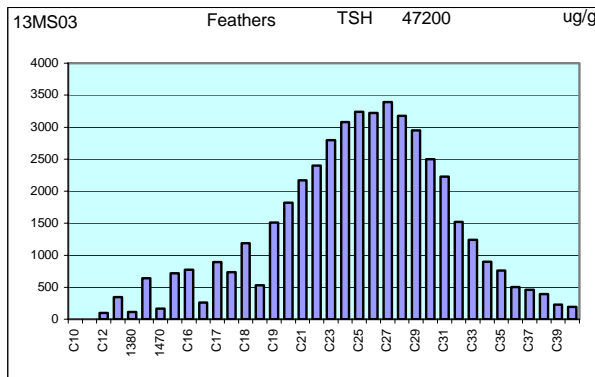
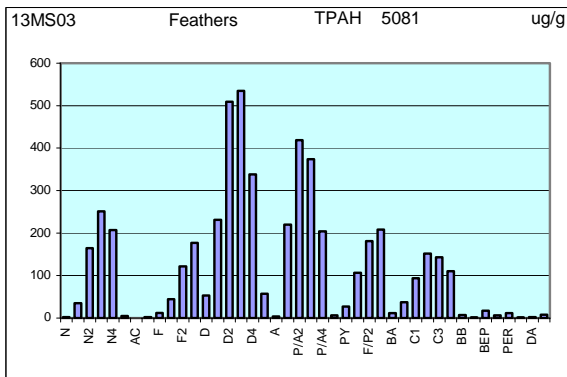
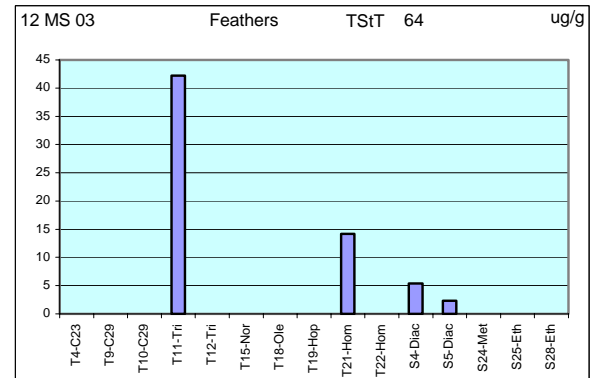
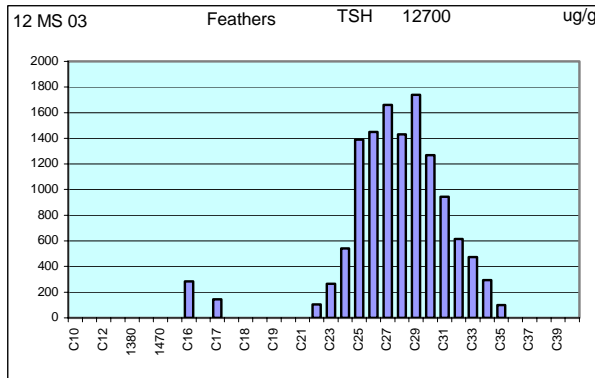
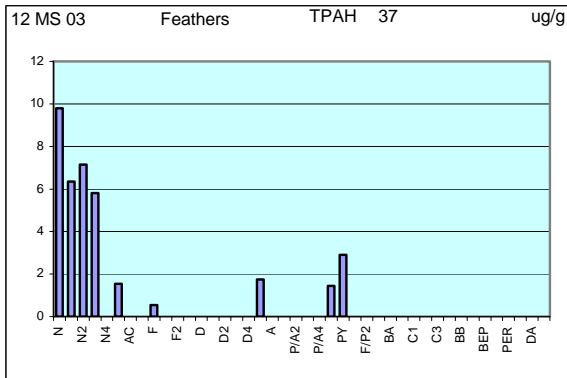
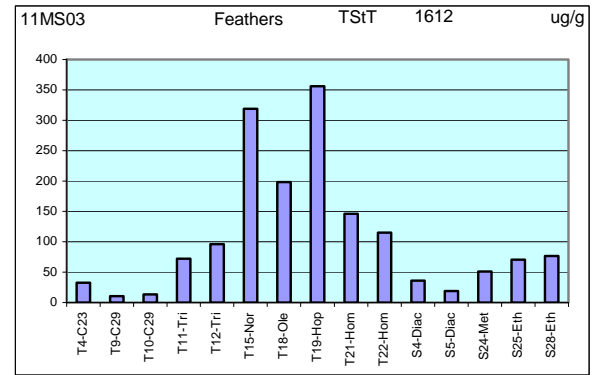
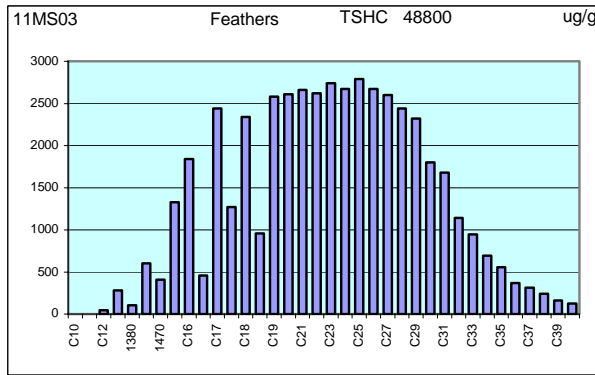
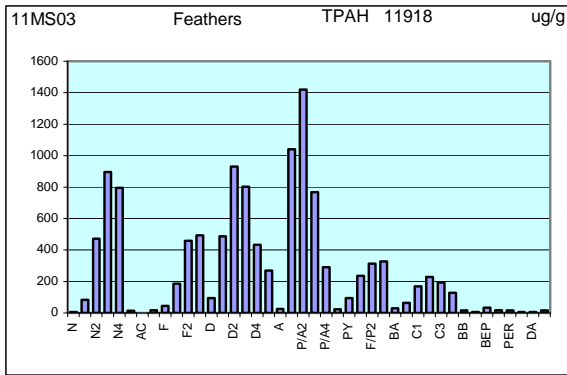


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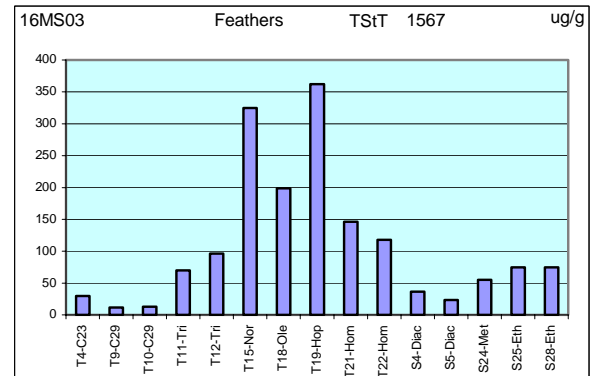
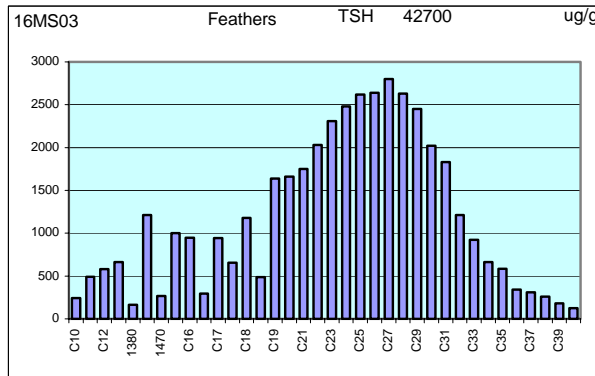
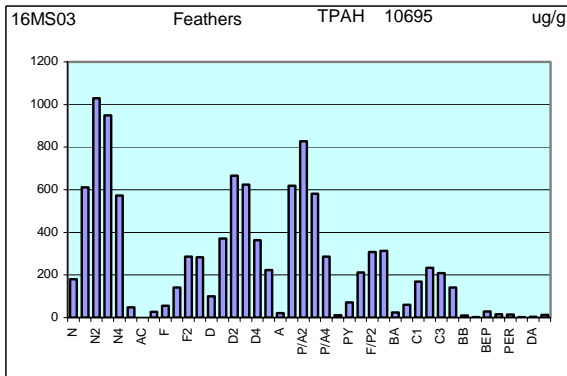
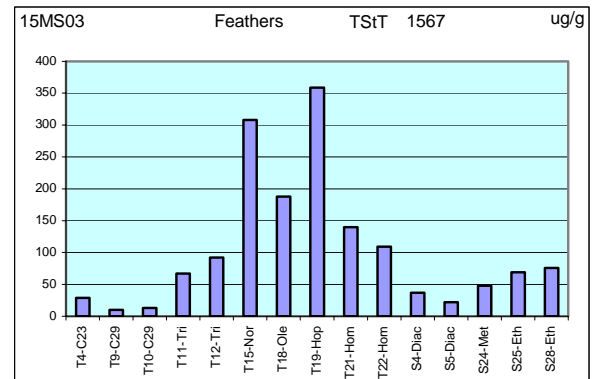
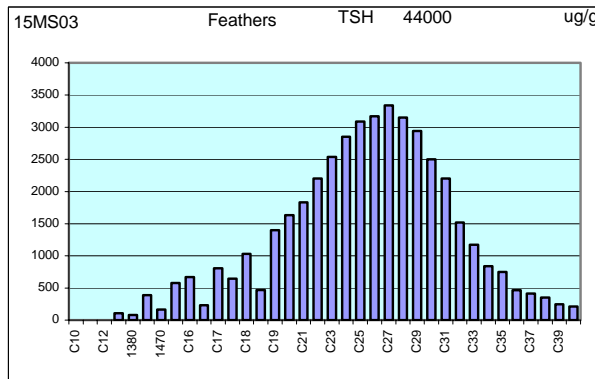
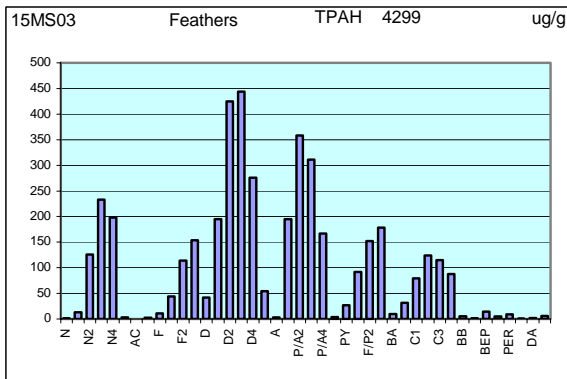
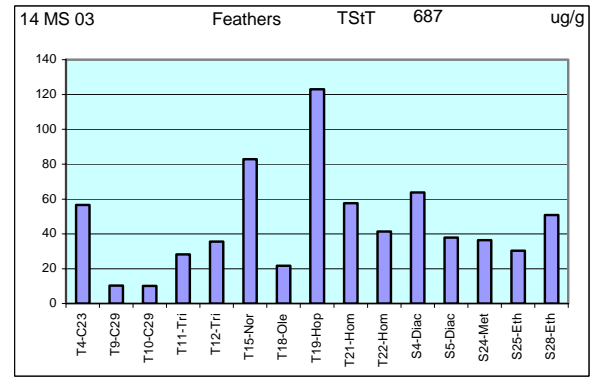
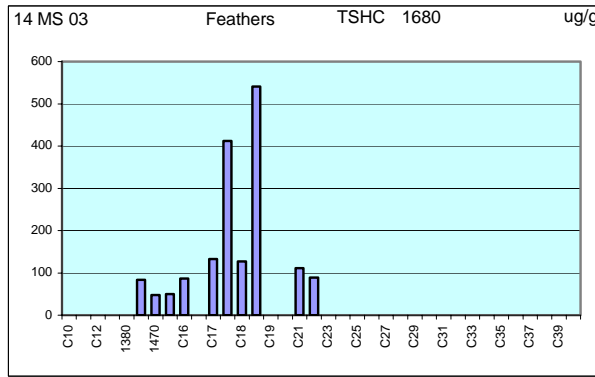
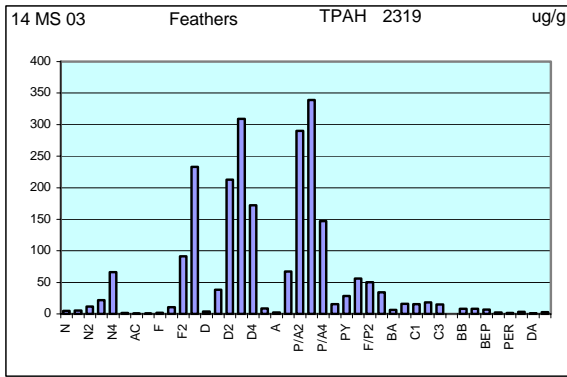




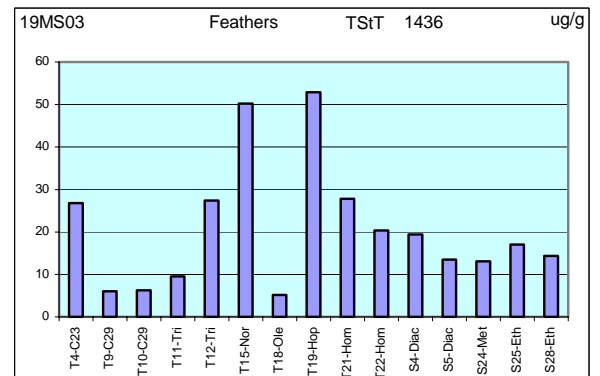
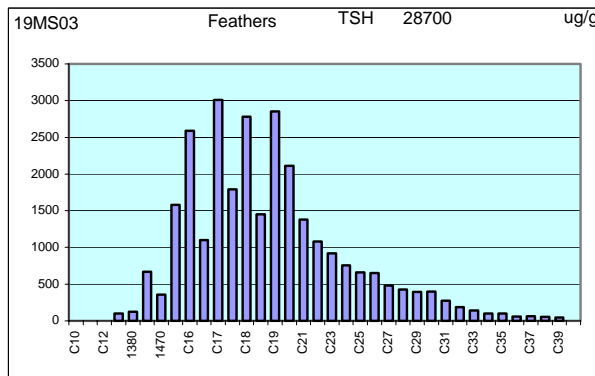
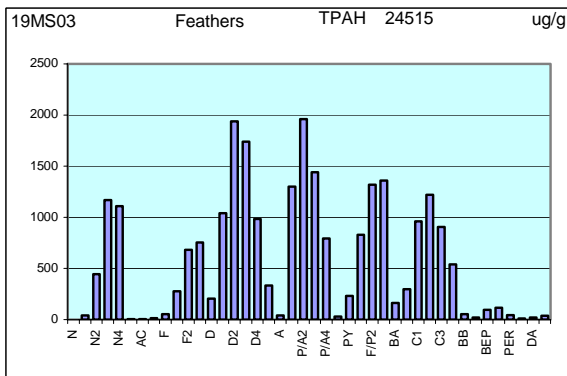
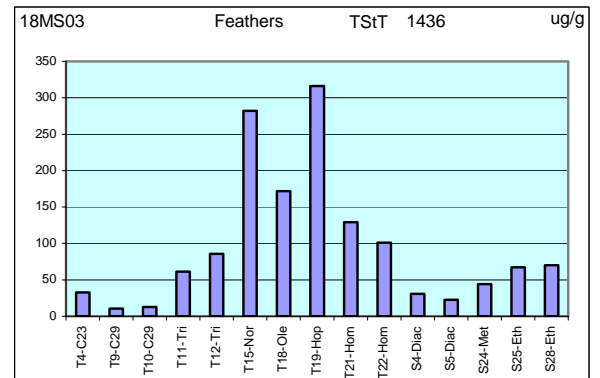
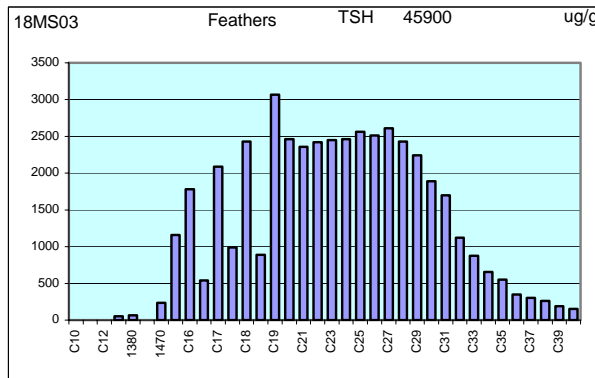
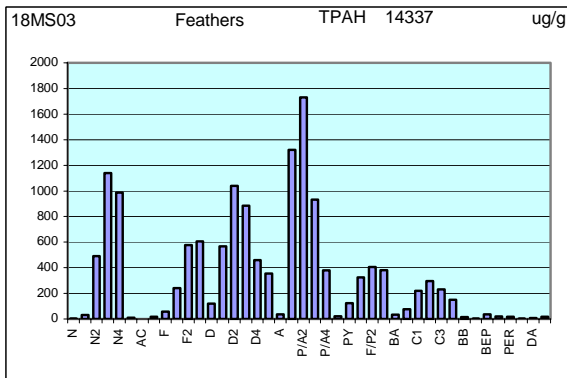
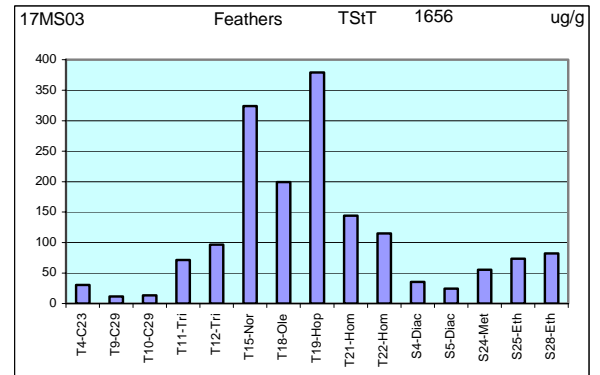
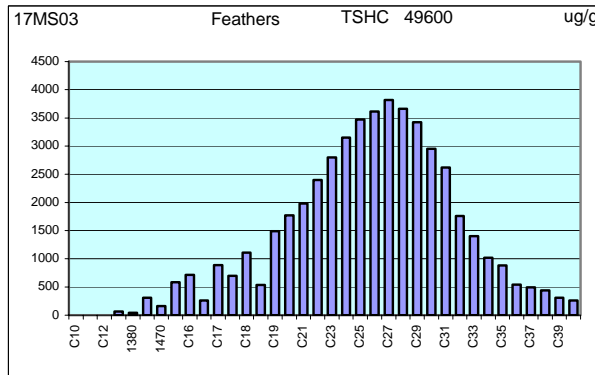
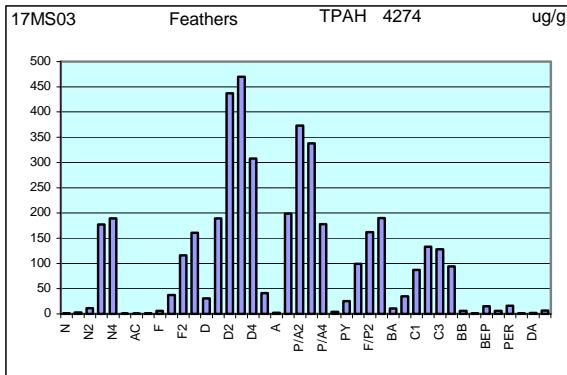
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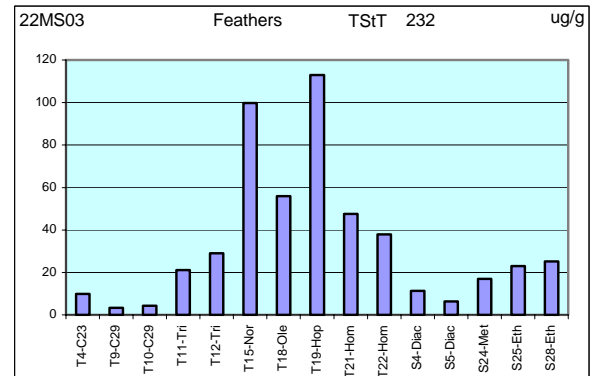
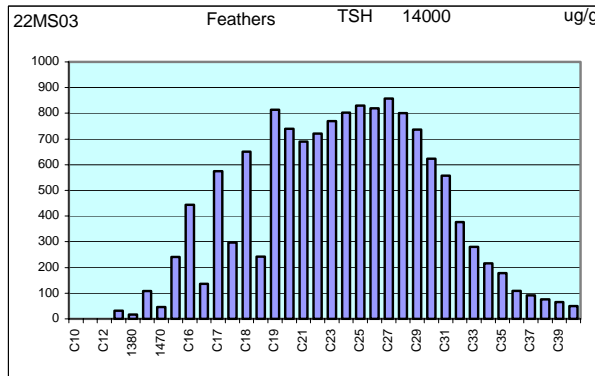
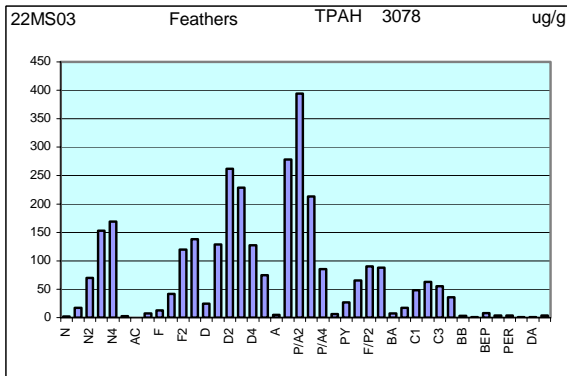
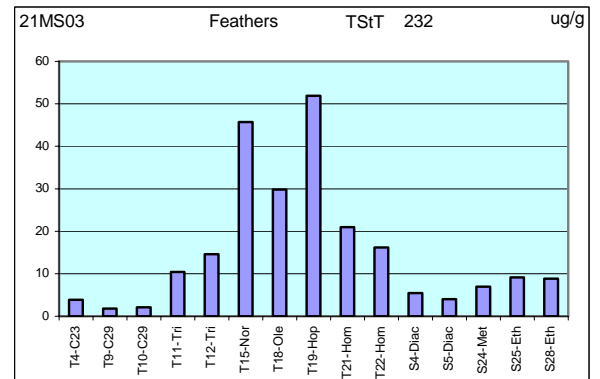
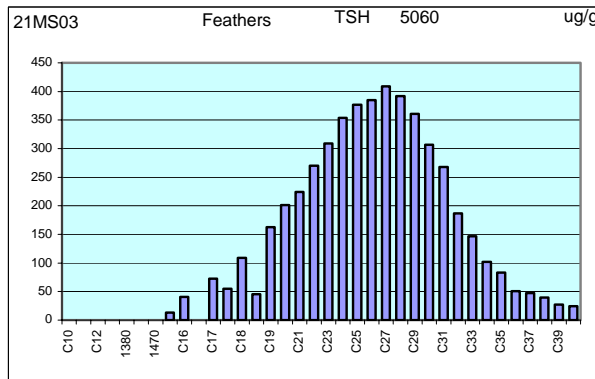
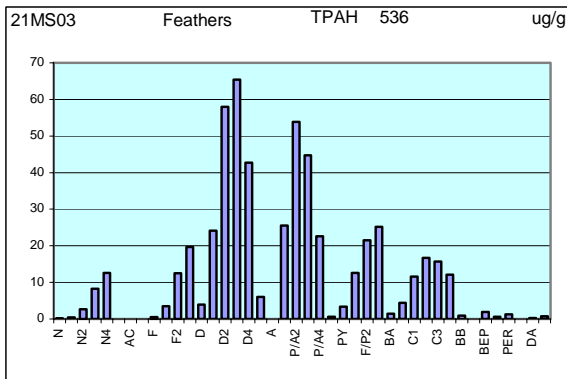
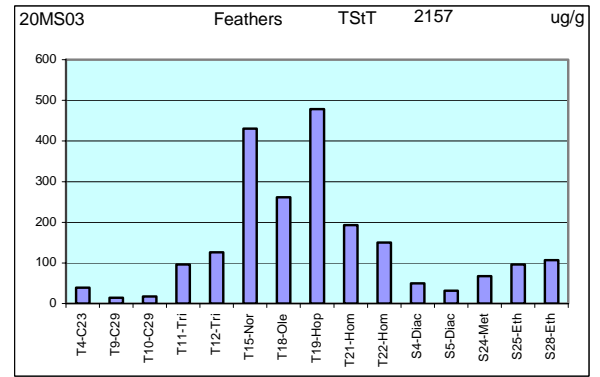
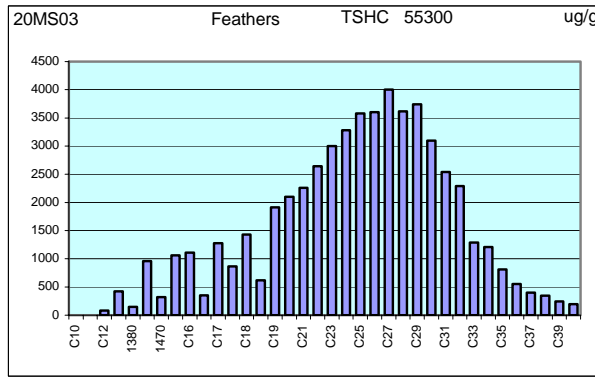
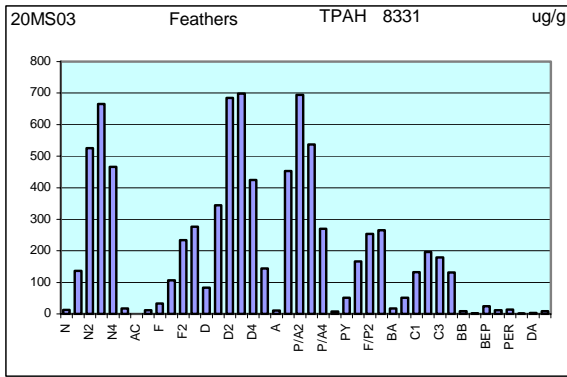
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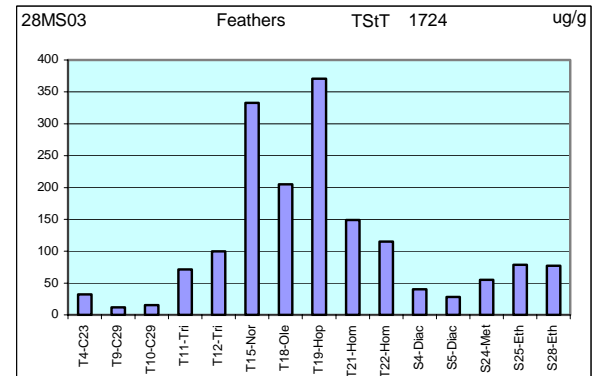
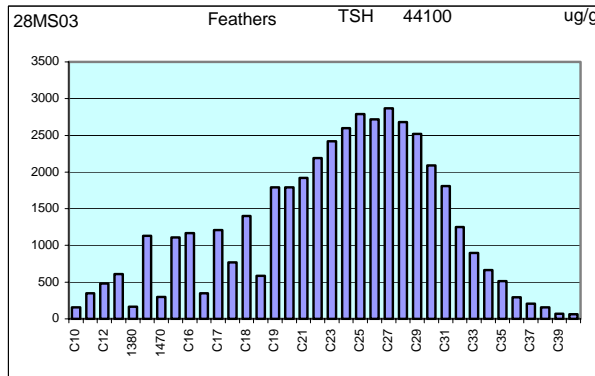
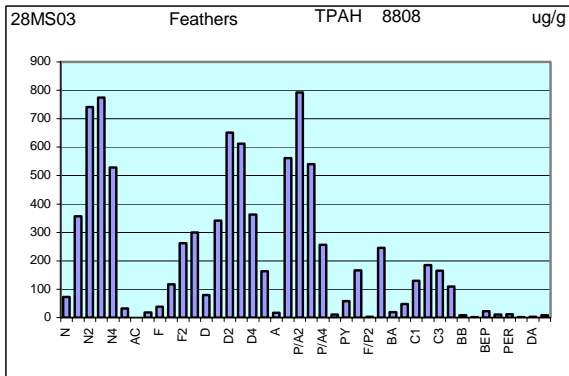
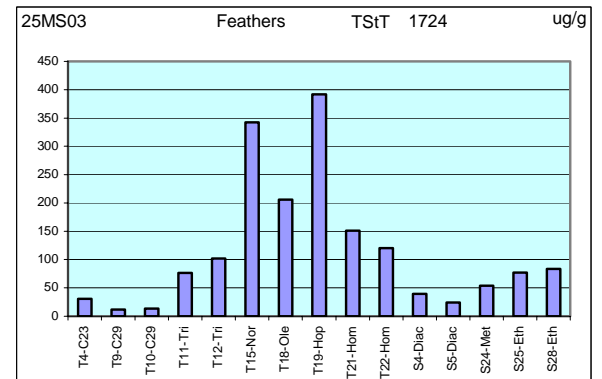
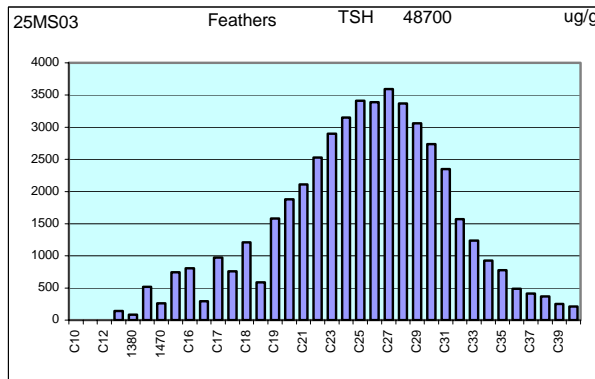
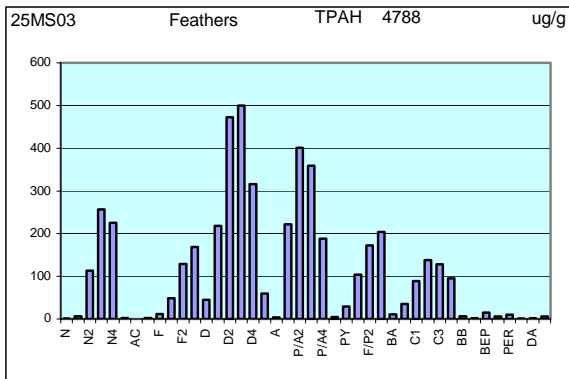
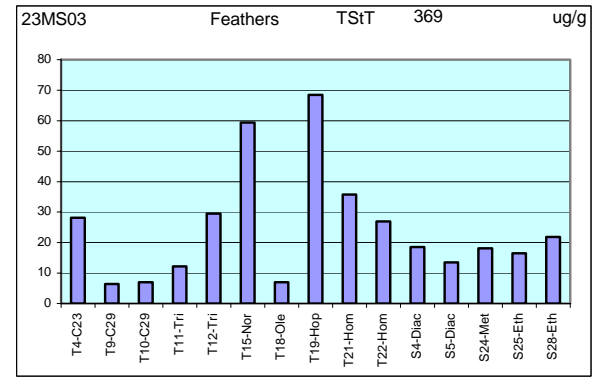
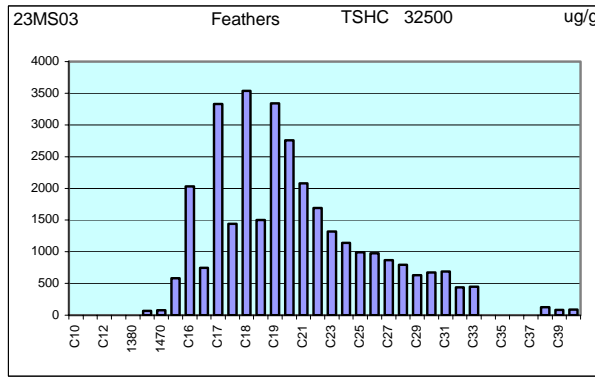
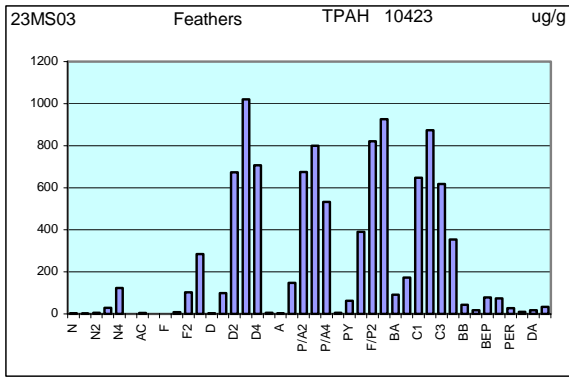
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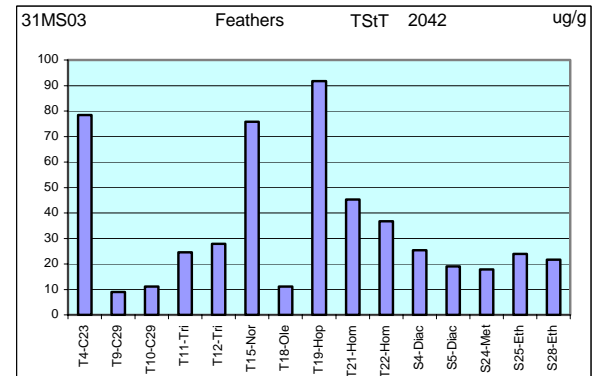
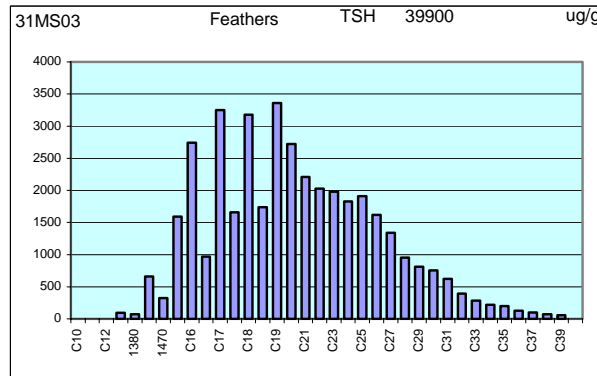
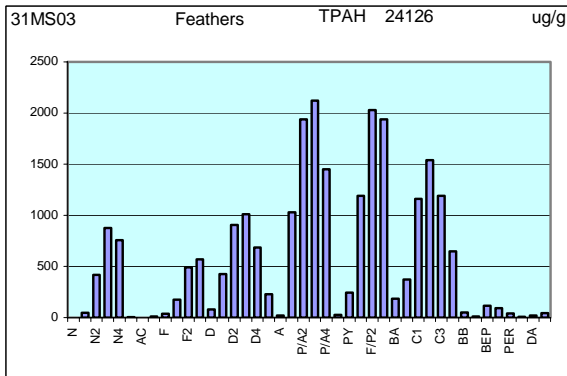
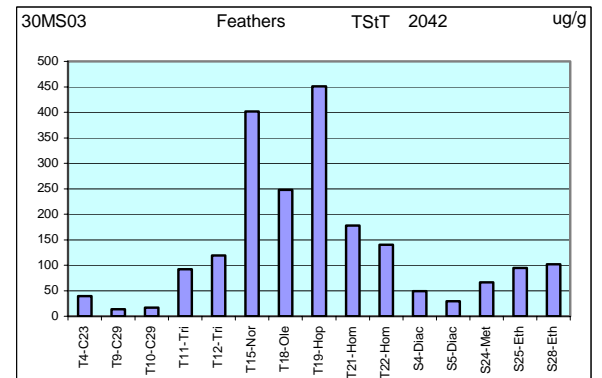
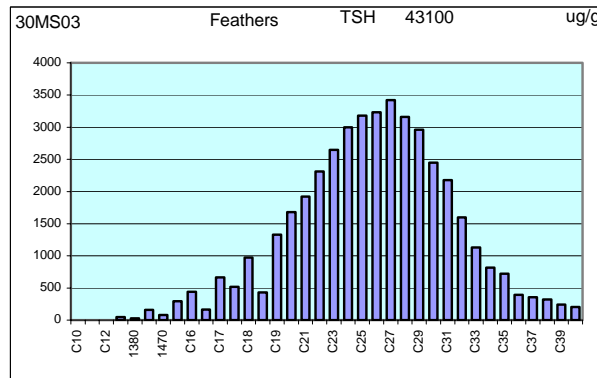
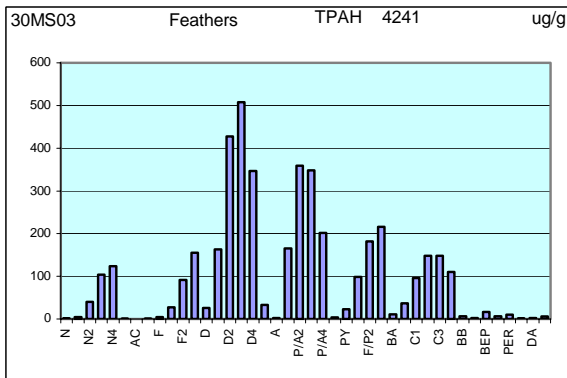
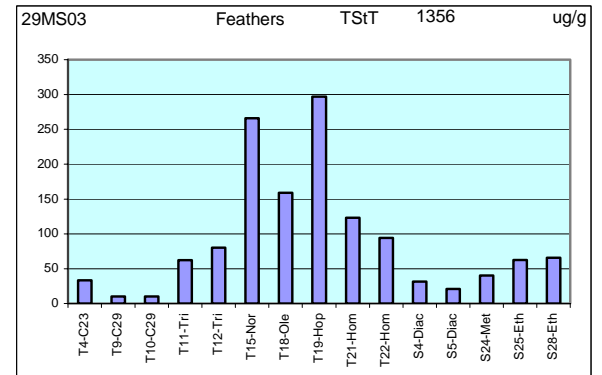
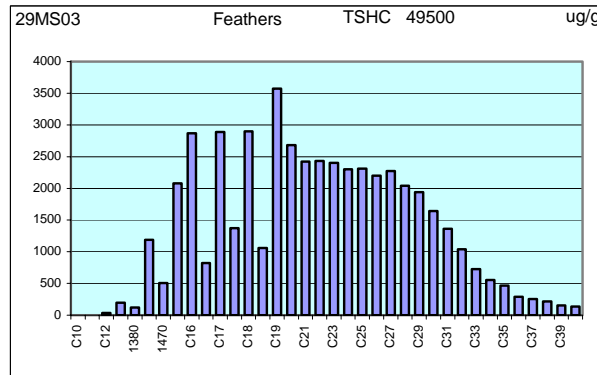
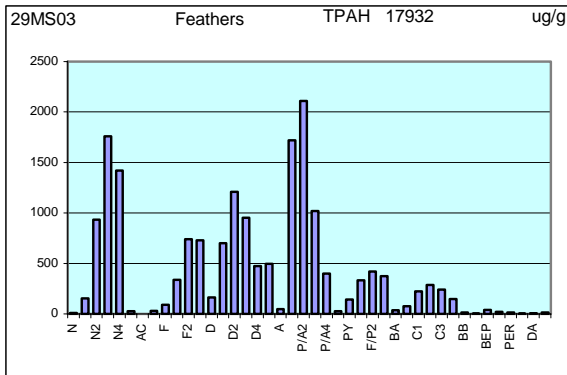
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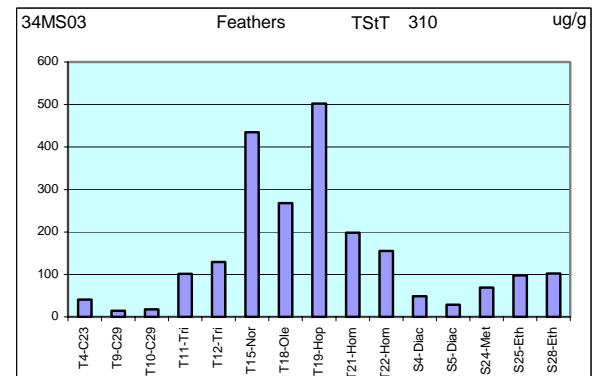
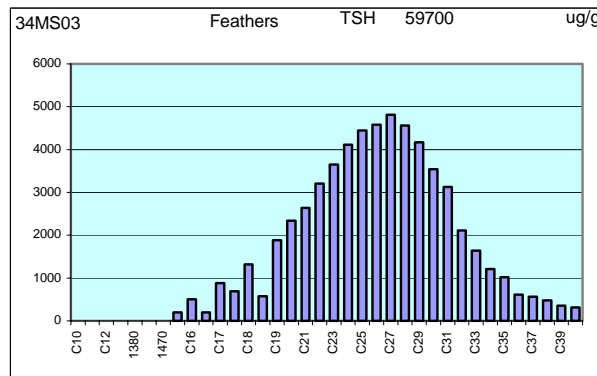
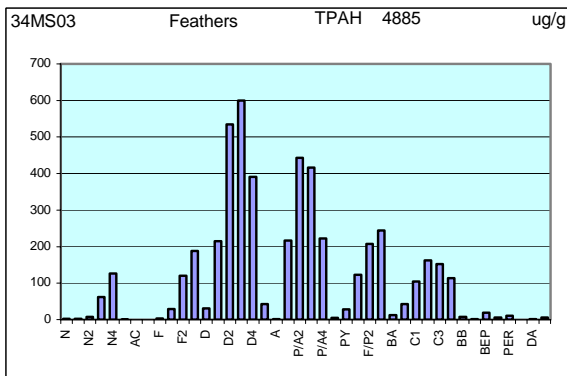
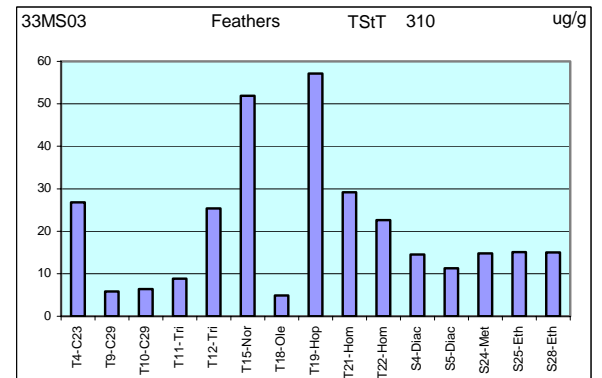
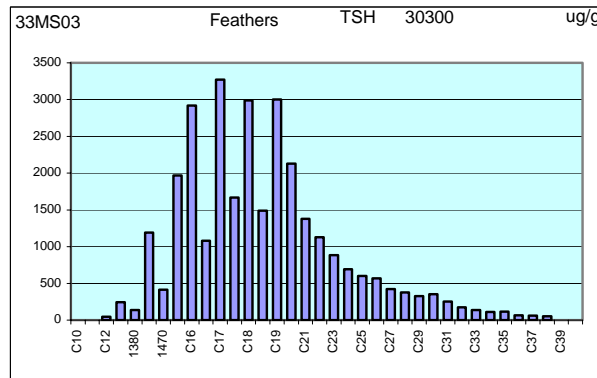
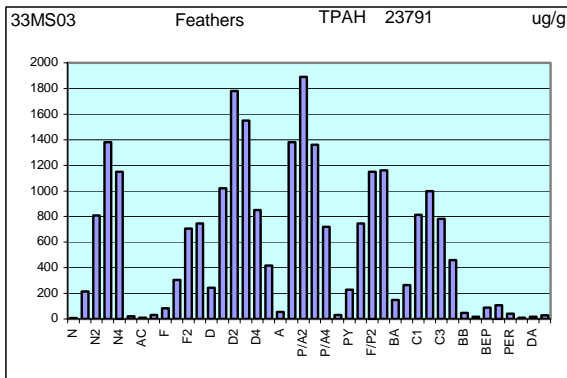
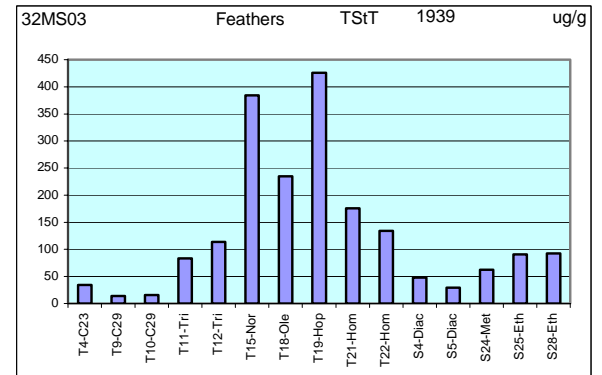
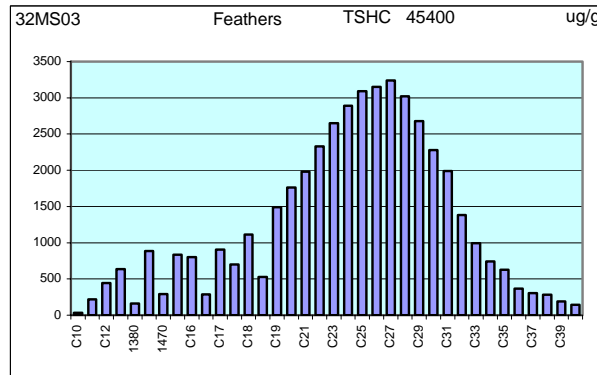
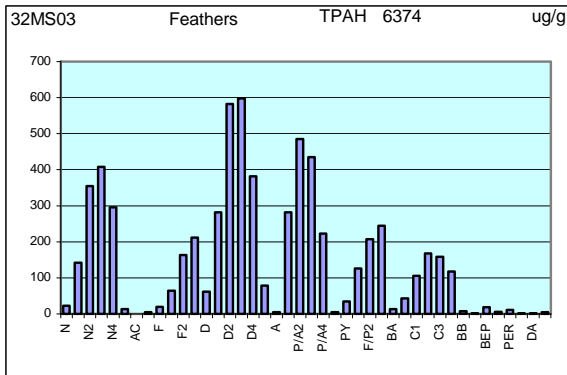
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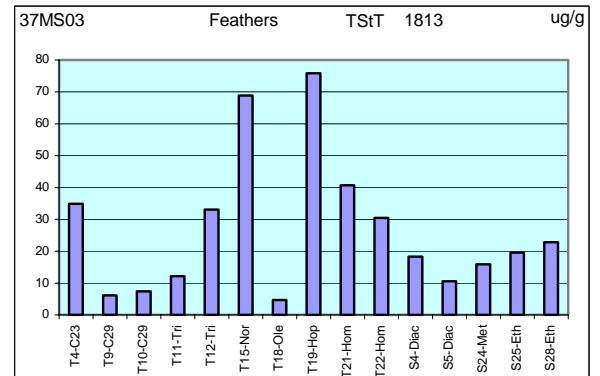
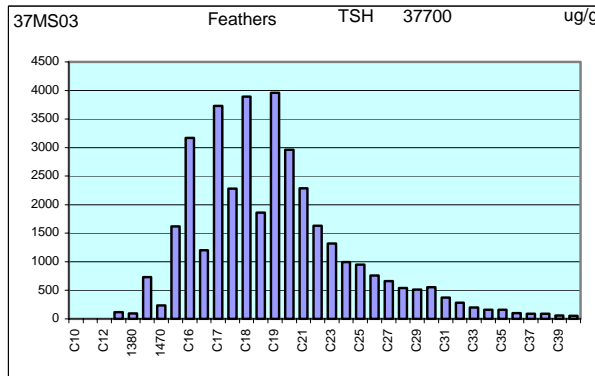
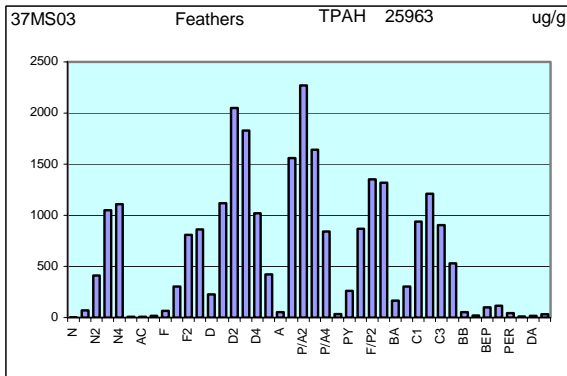
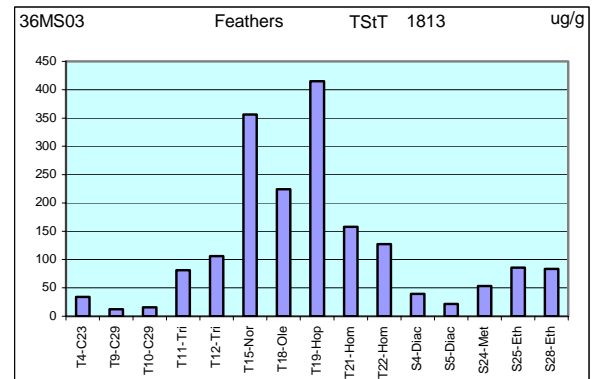
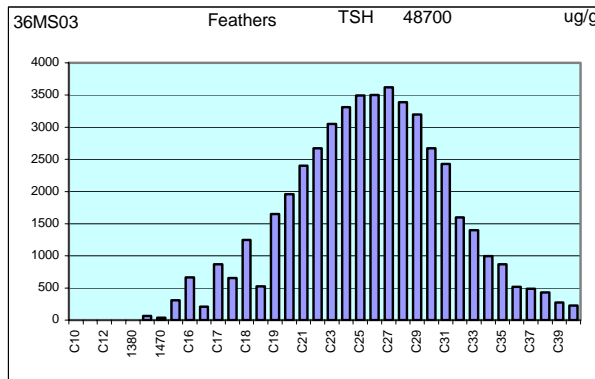
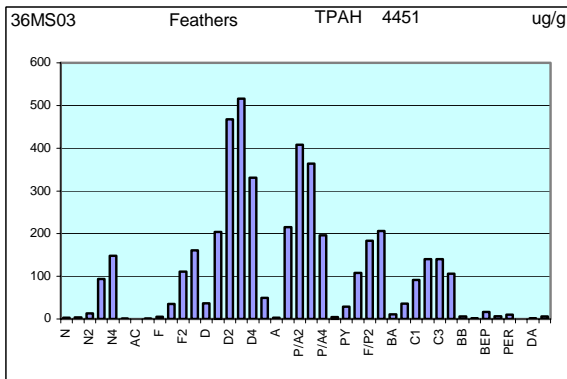
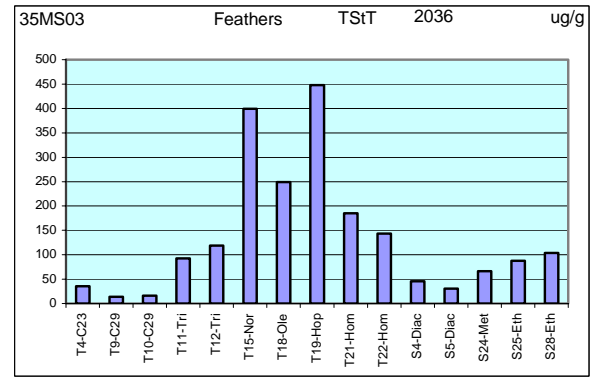
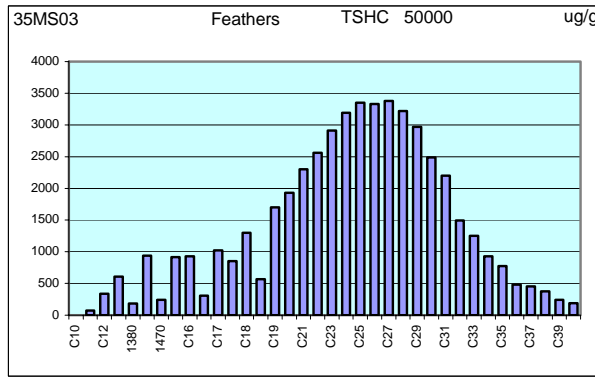
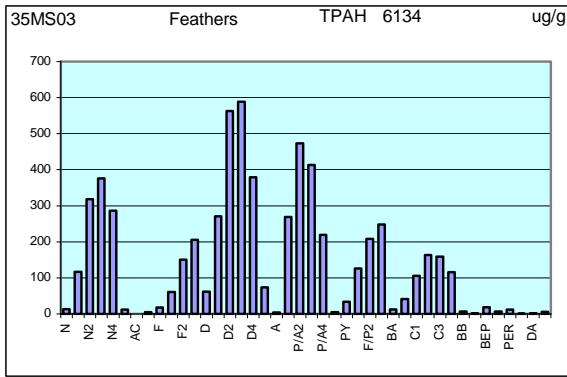
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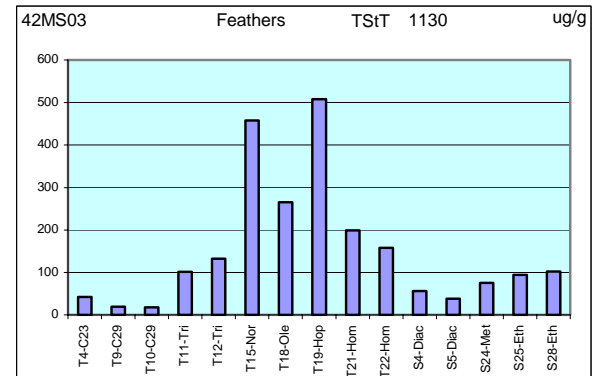
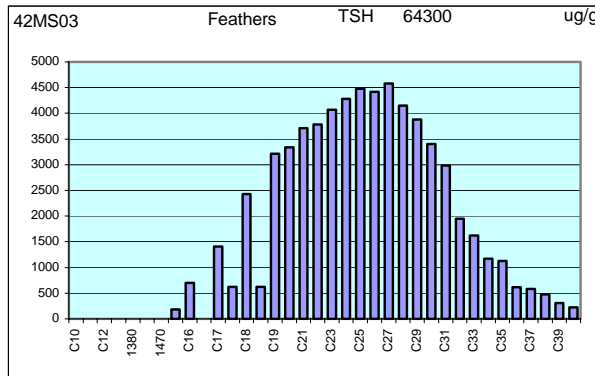
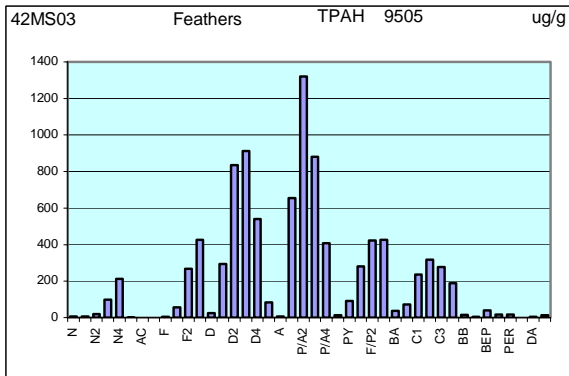
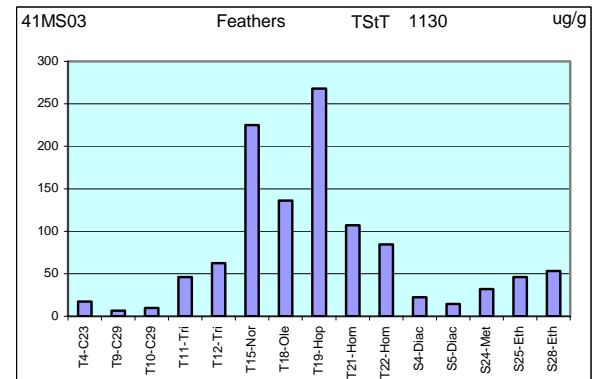
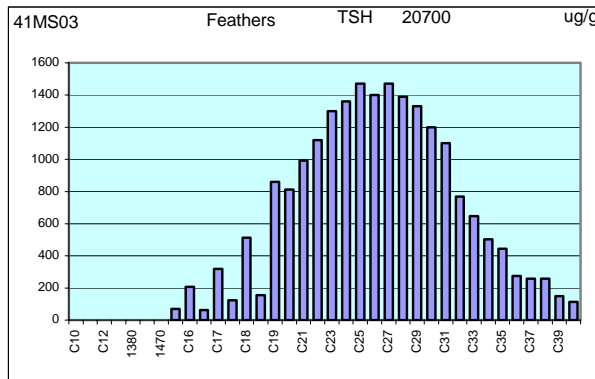
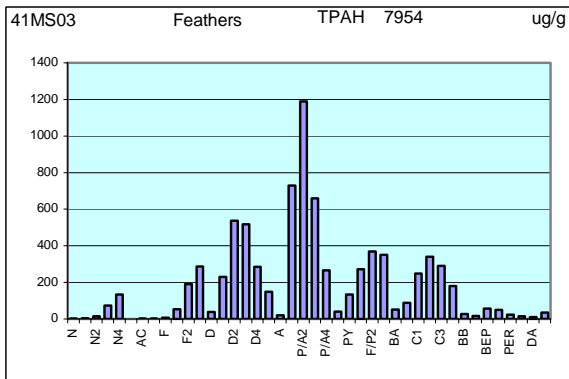
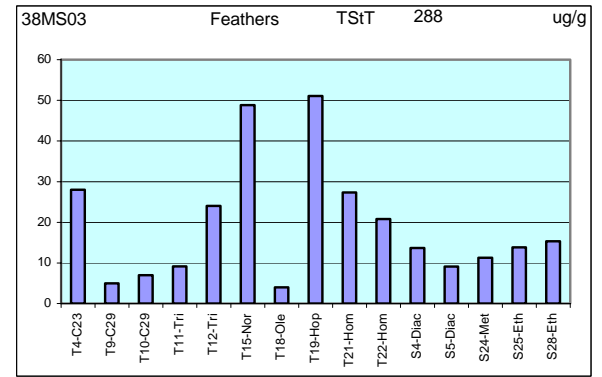
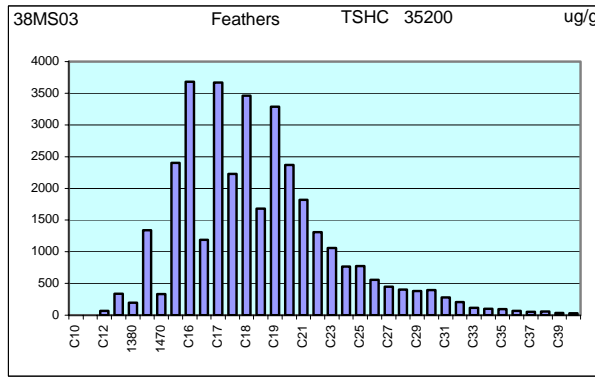
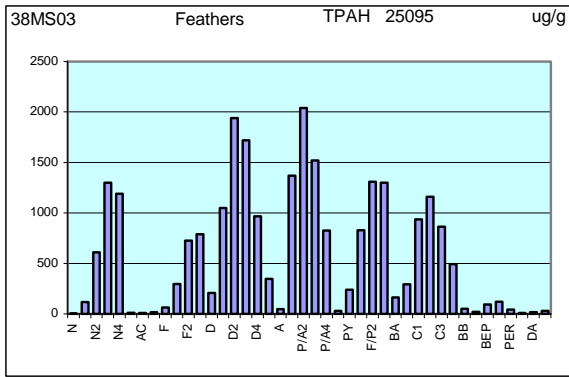


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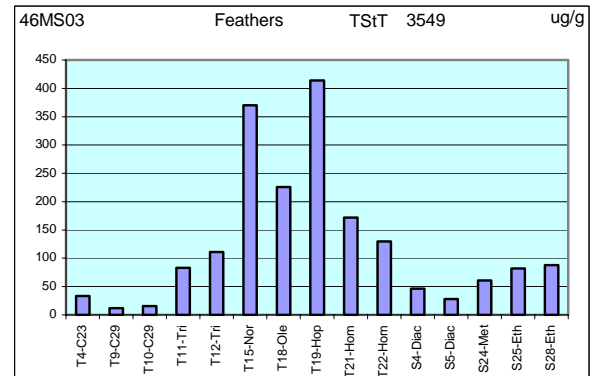
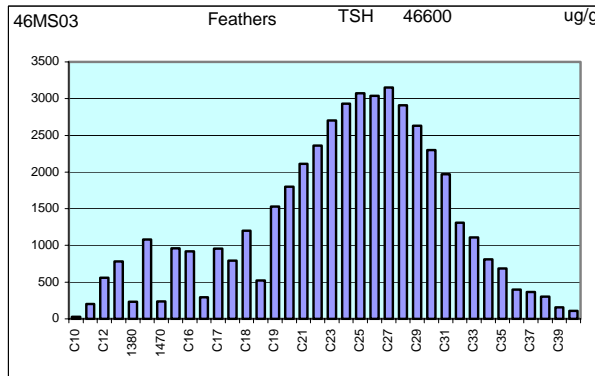
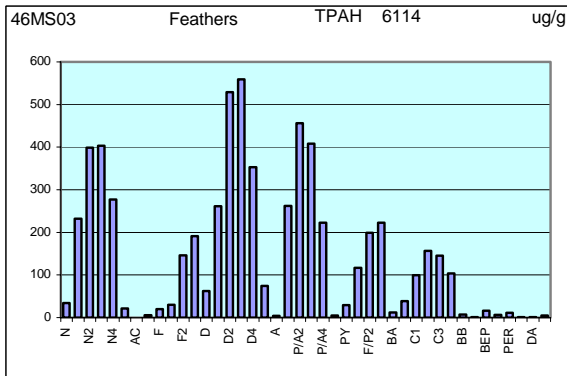
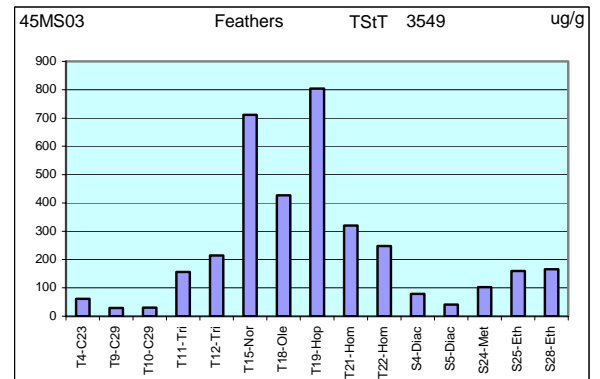
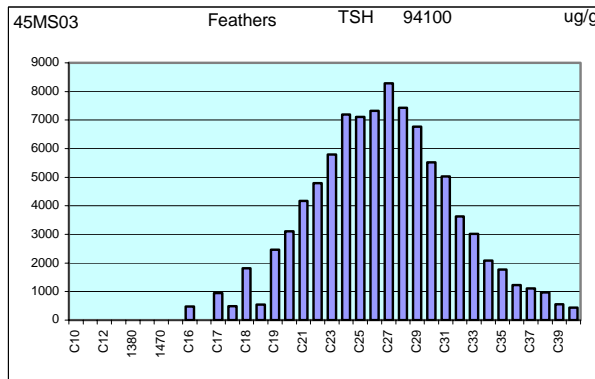
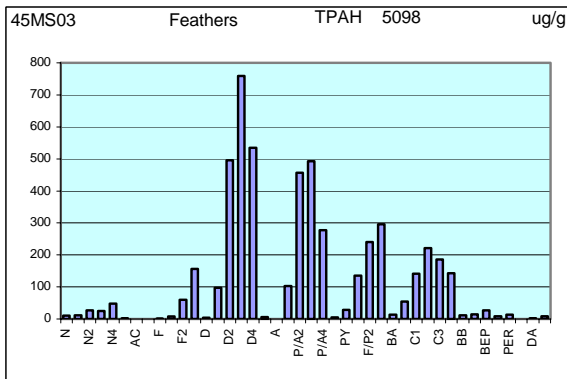
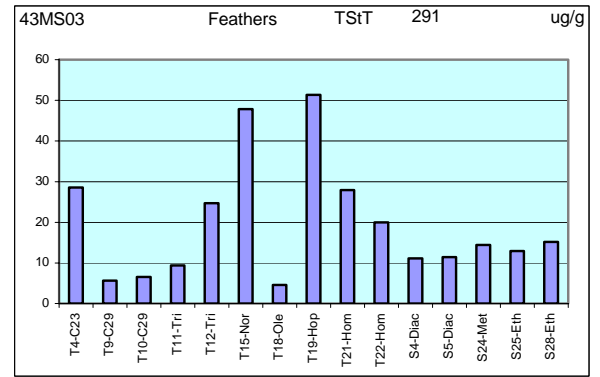
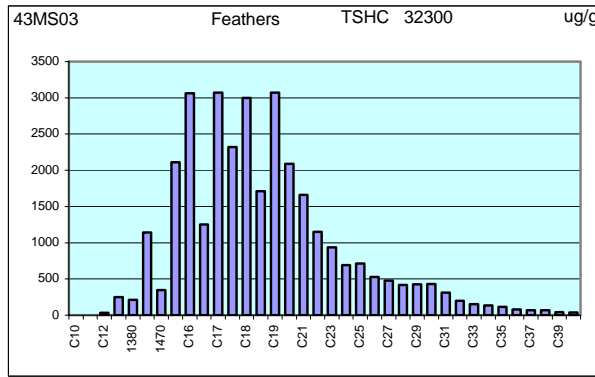
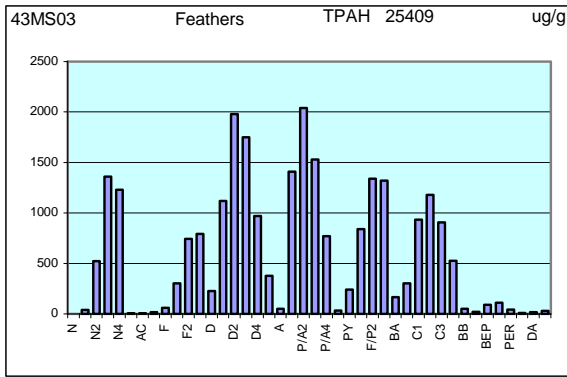




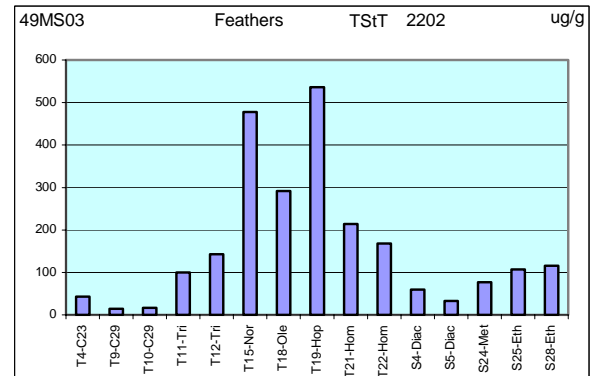
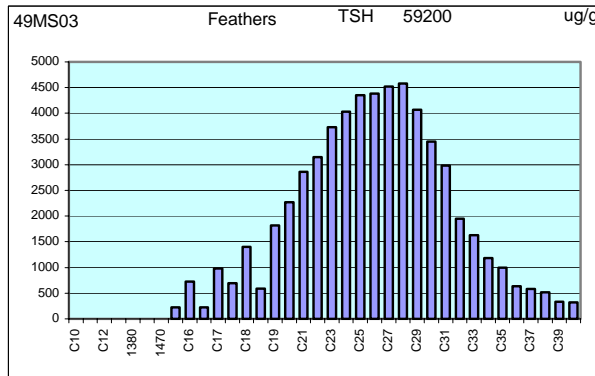
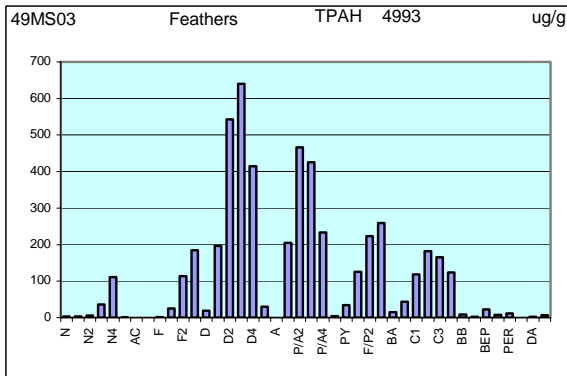
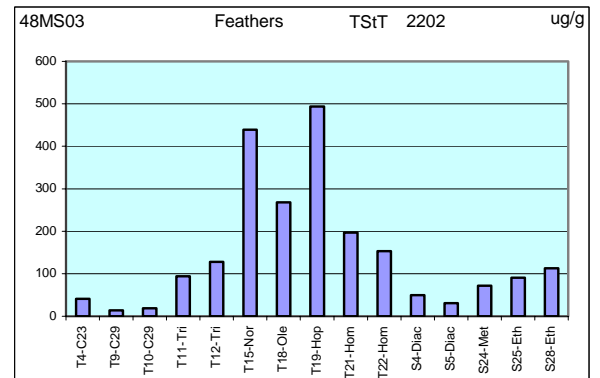
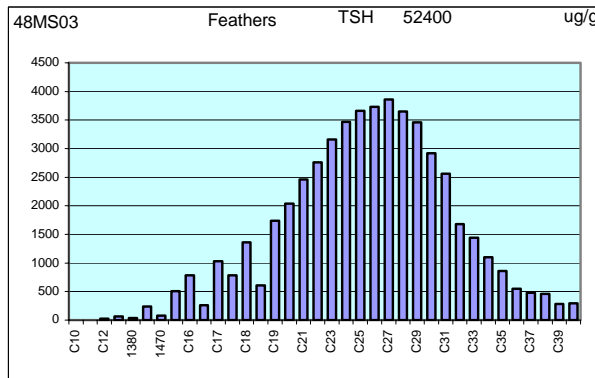
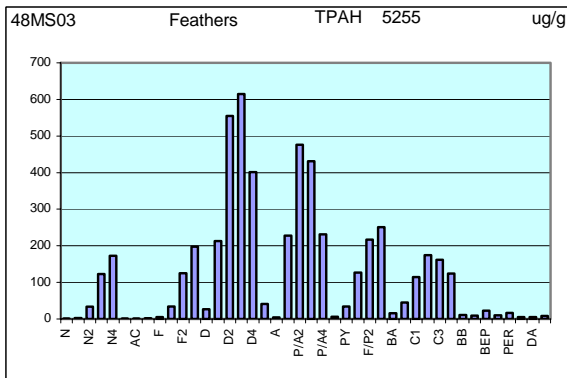
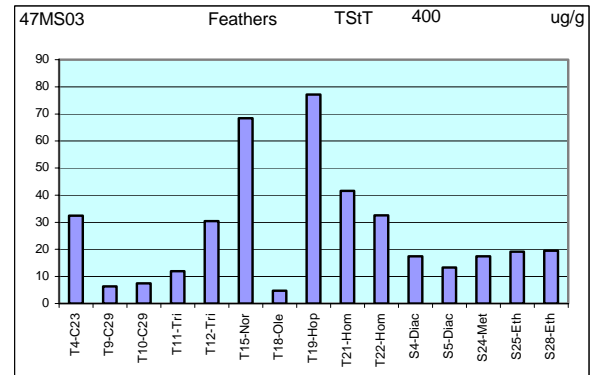
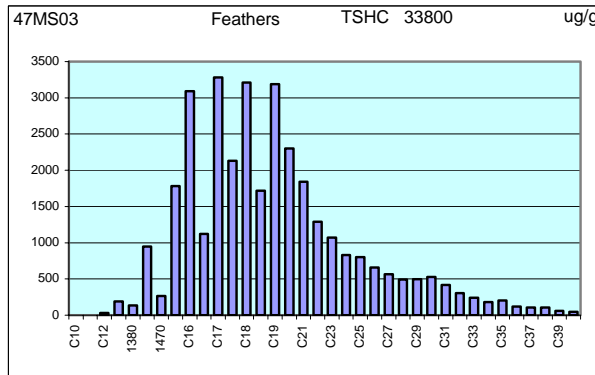
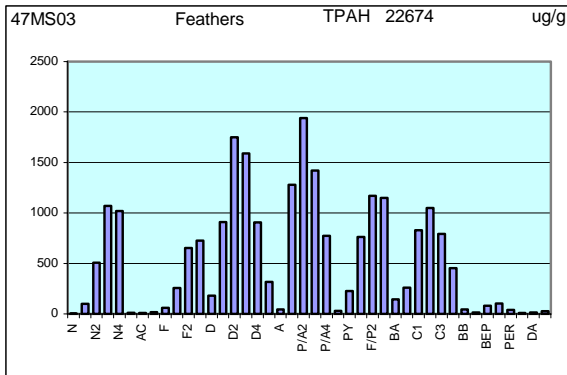
**WORK PRODUCT – ATTORNEY CLIENT PRIVILEGED INFORMATION**



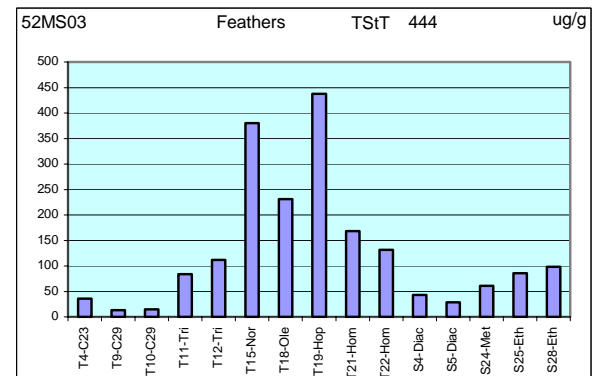
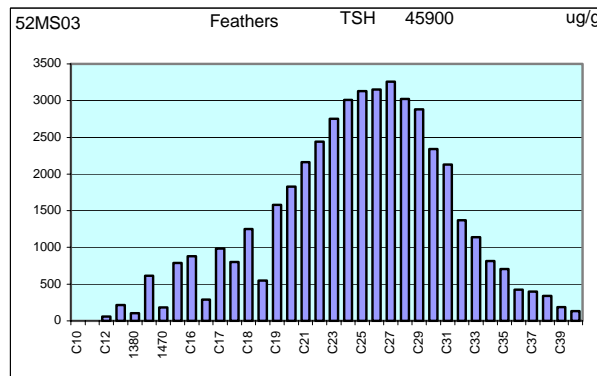
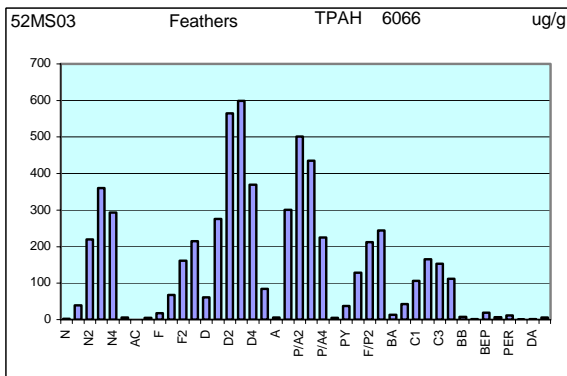
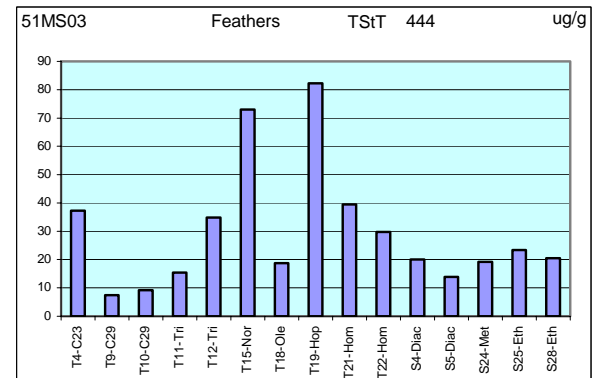
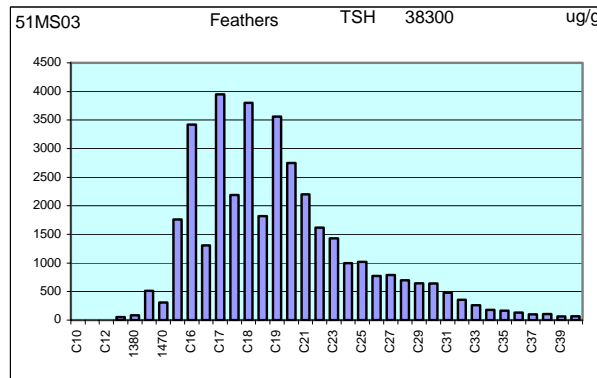
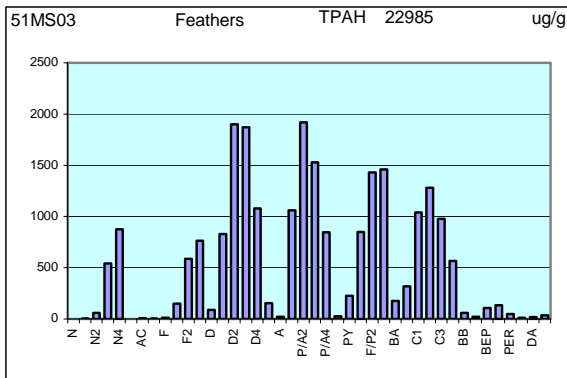
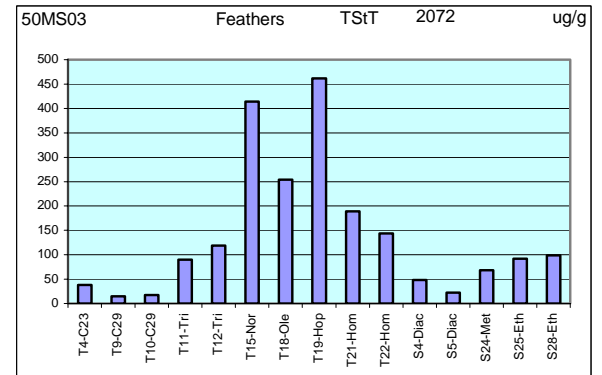
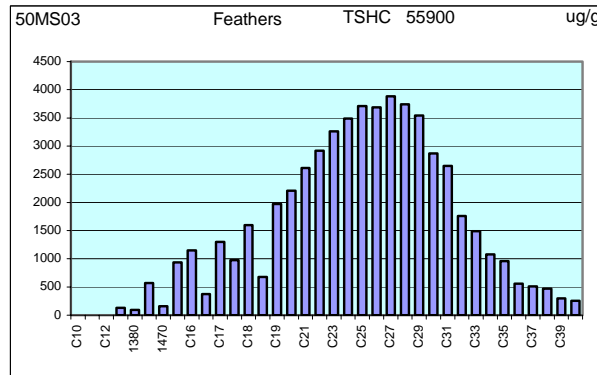
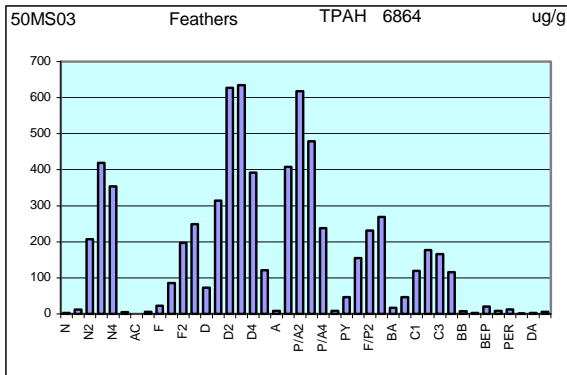
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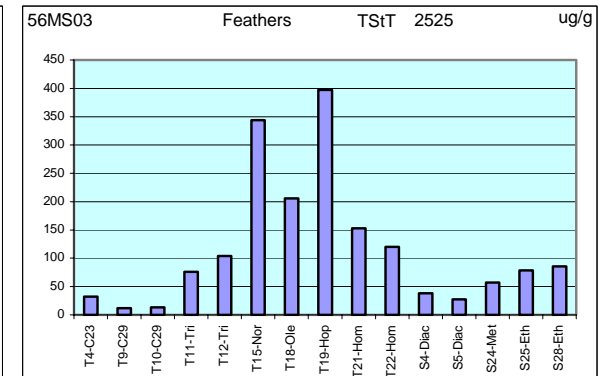
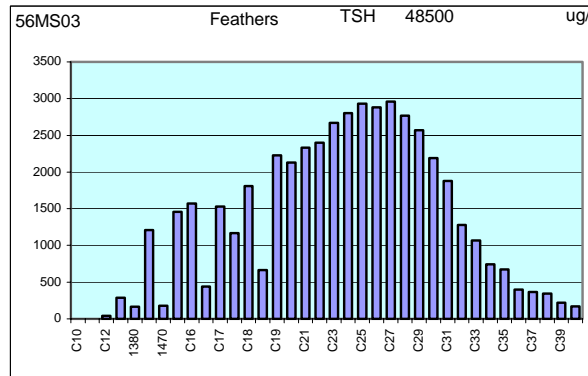
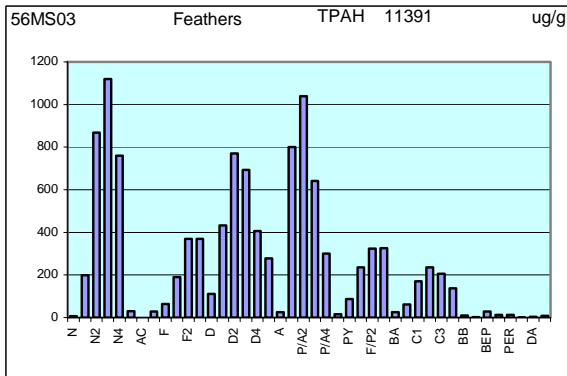
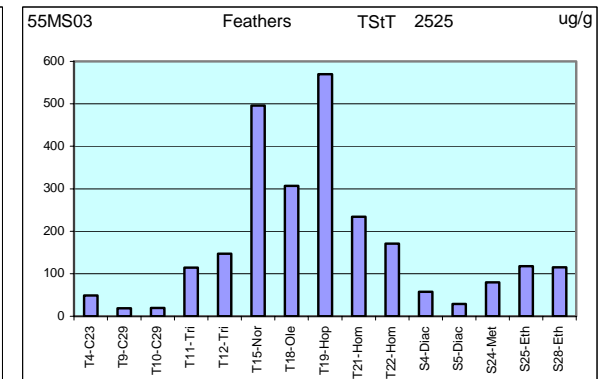
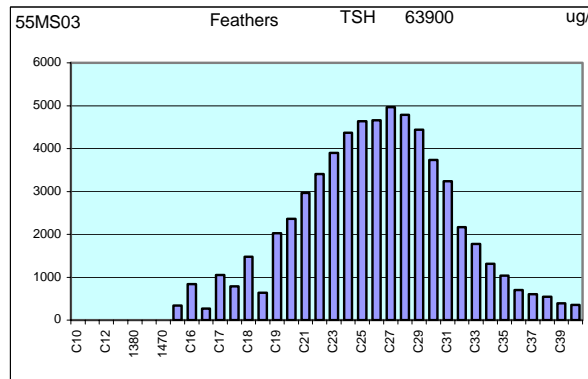
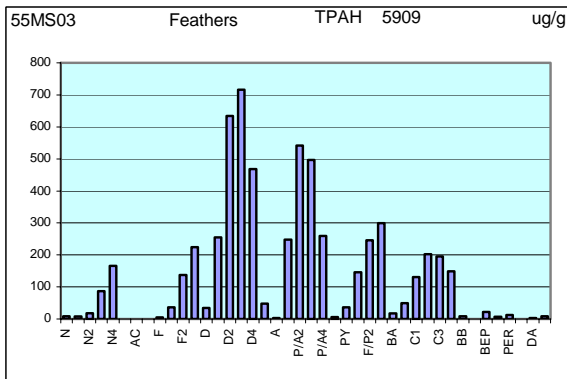
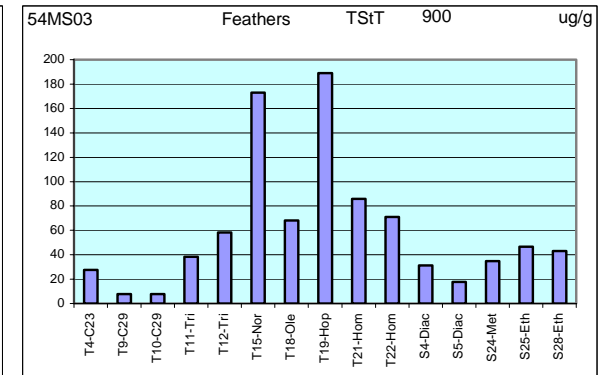
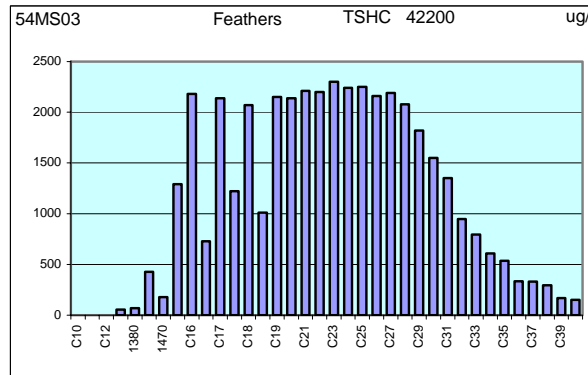
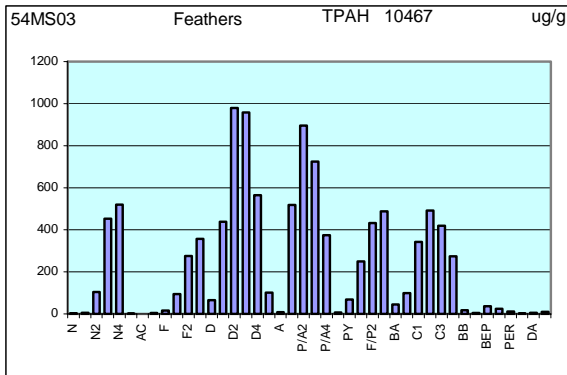
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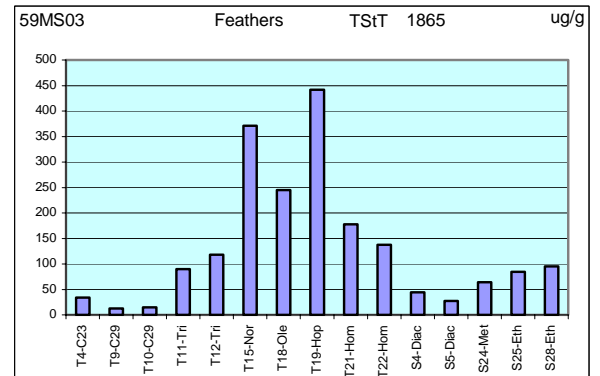
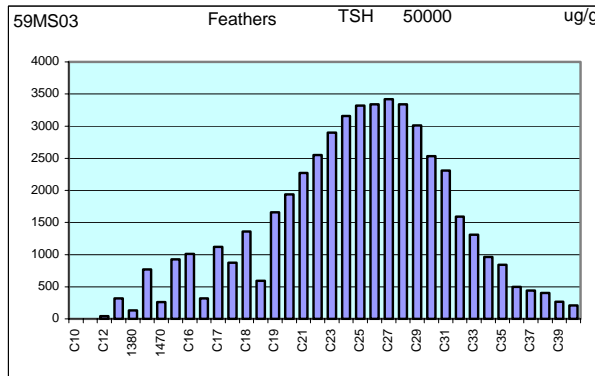
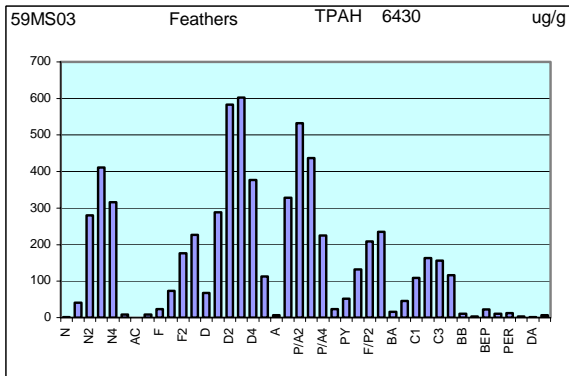
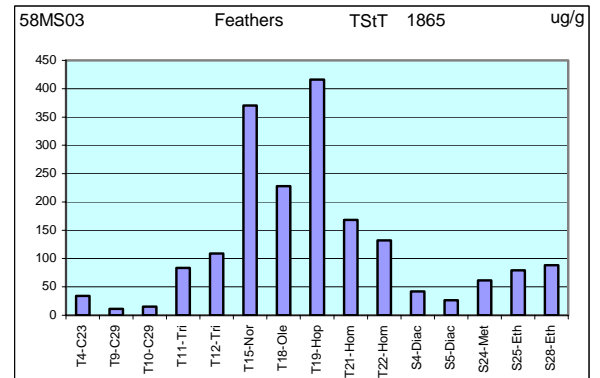
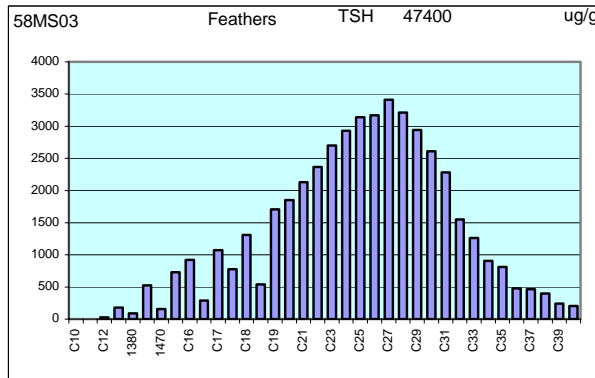
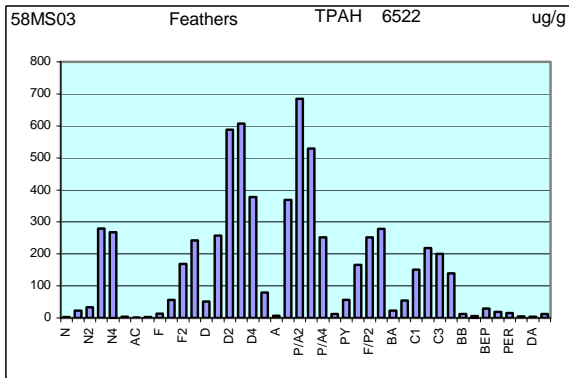
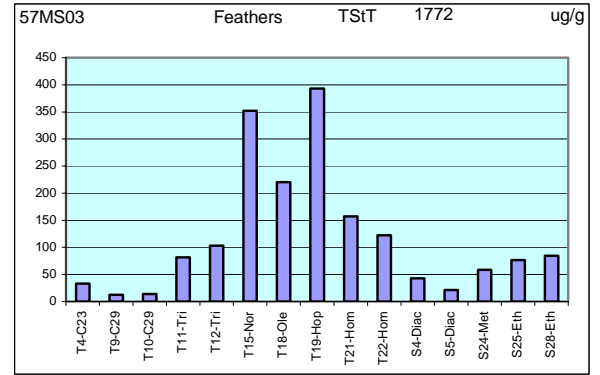
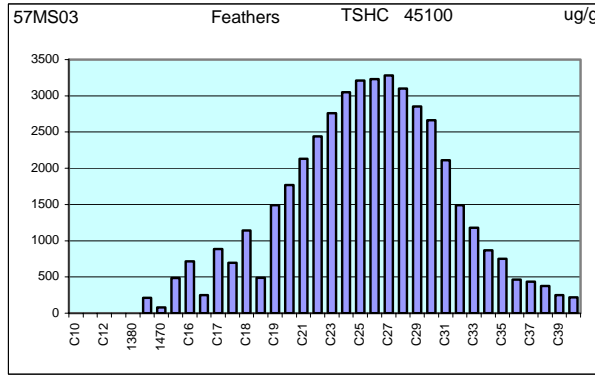
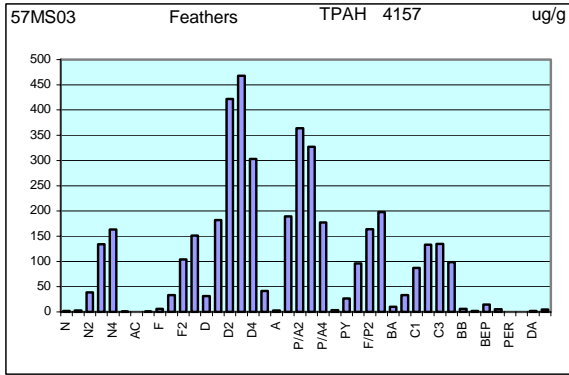
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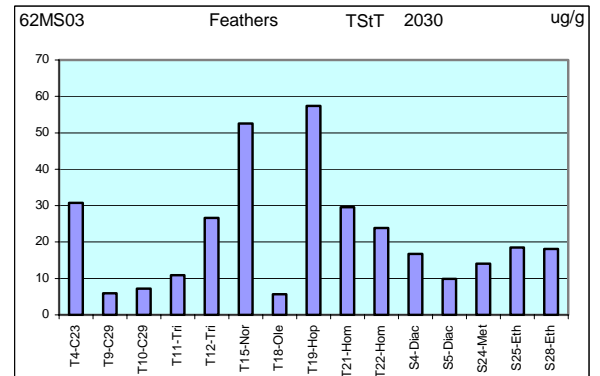
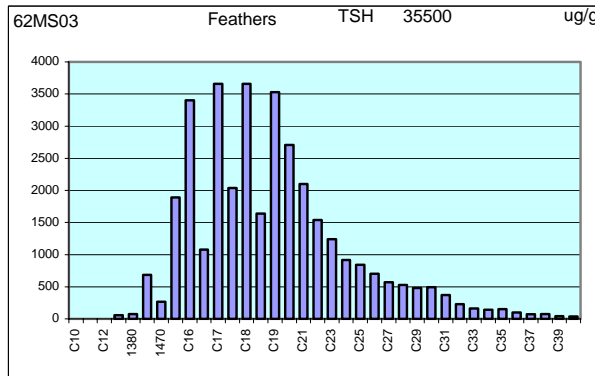
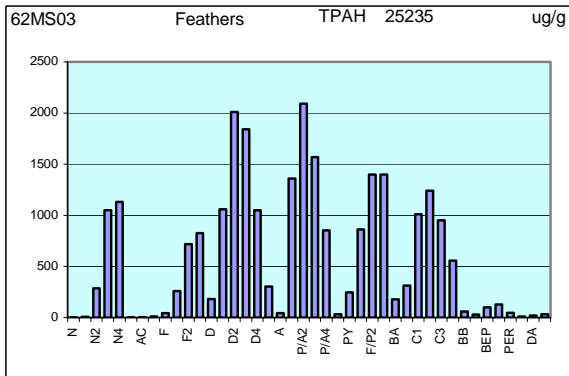
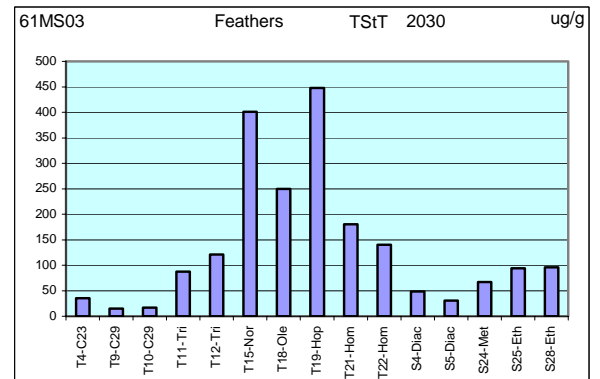
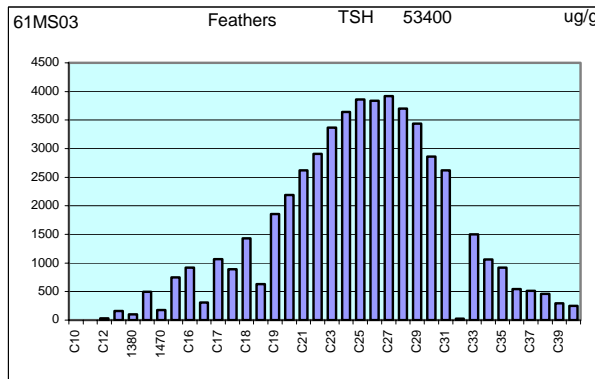
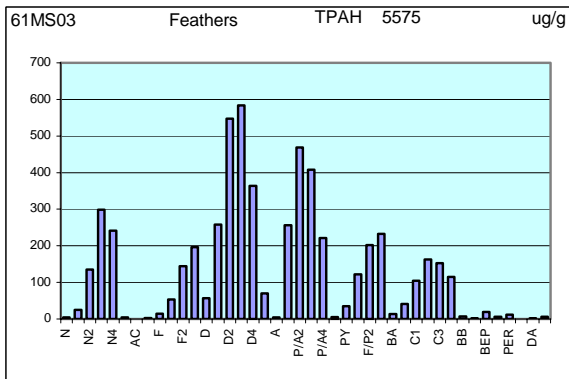
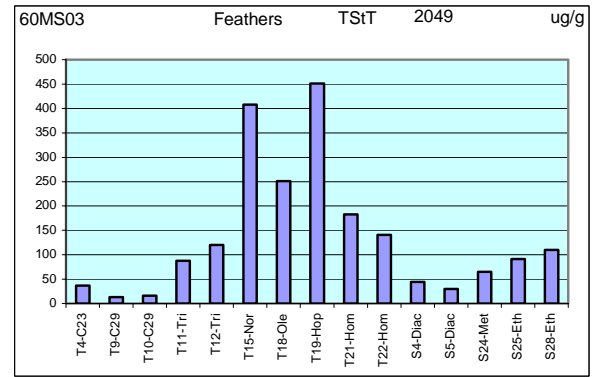
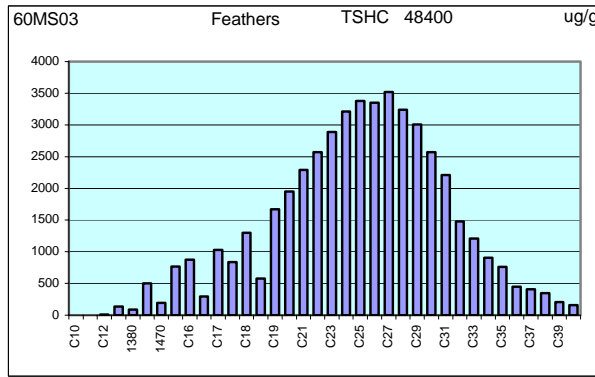
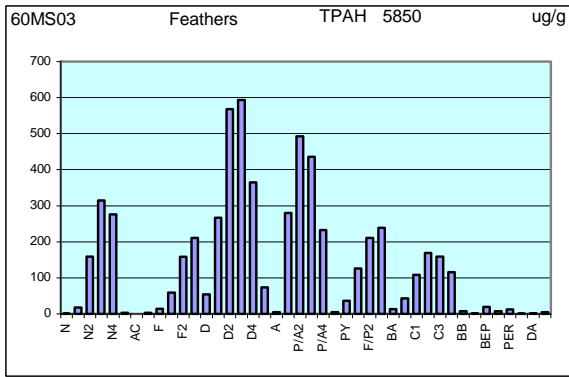
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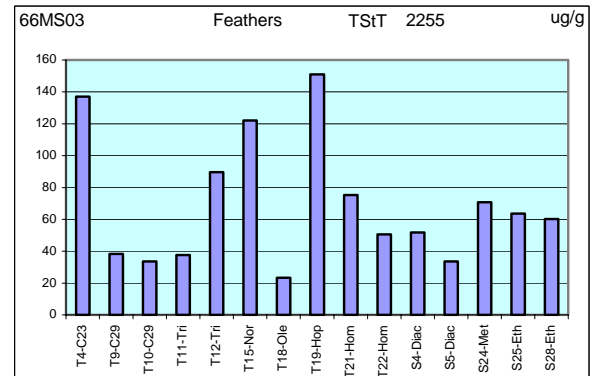
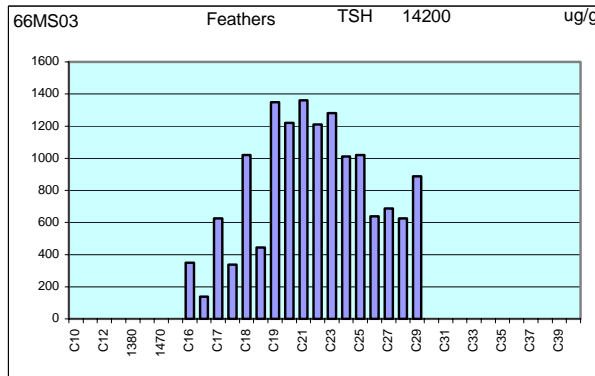
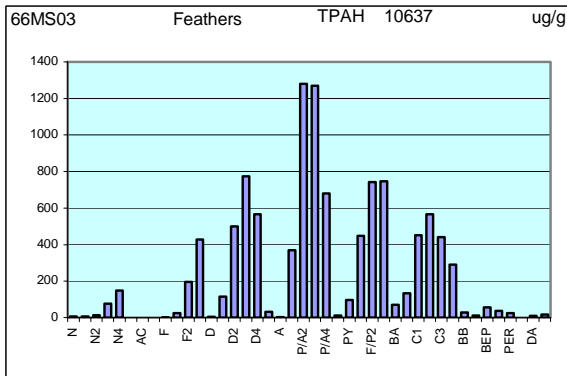
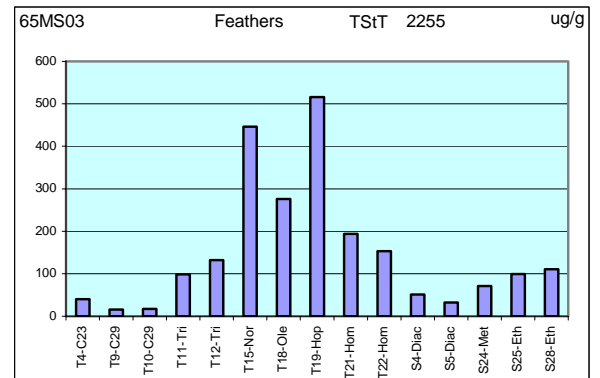
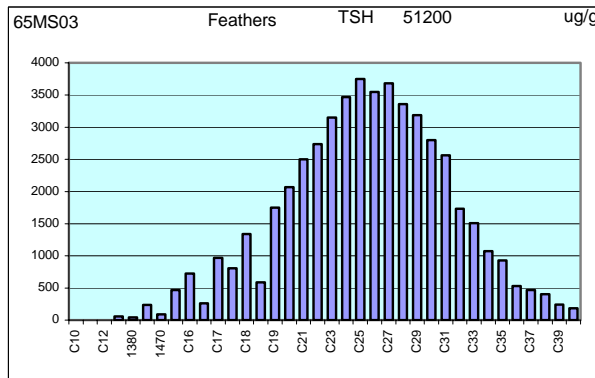
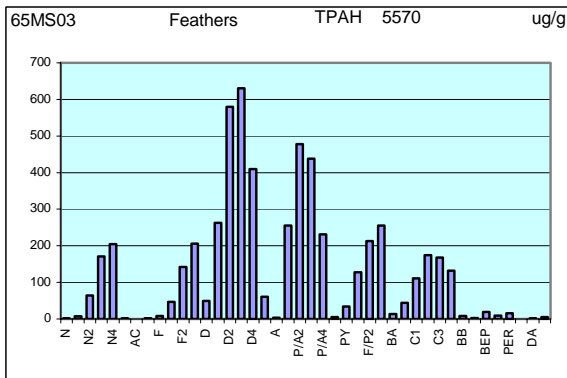
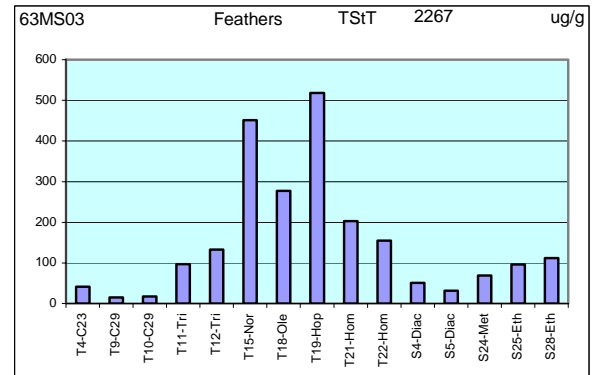
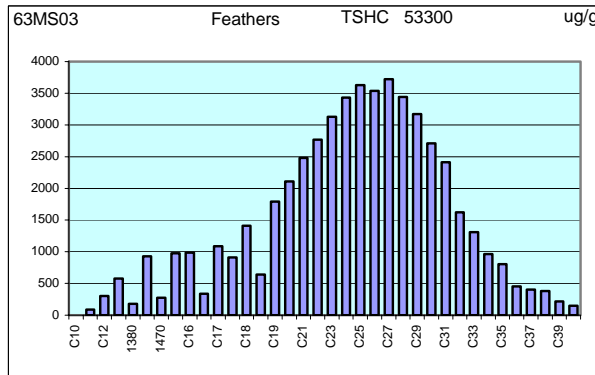
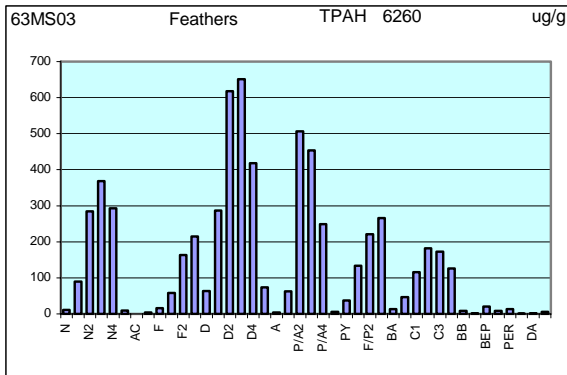
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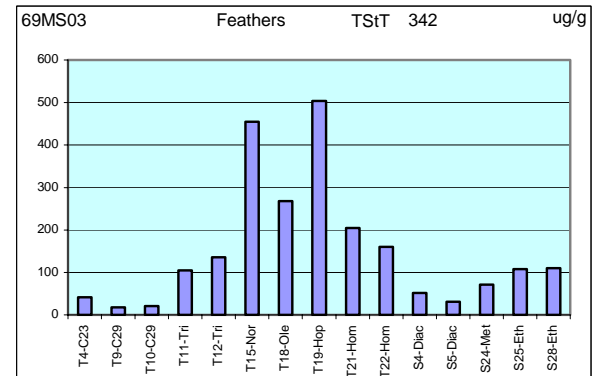
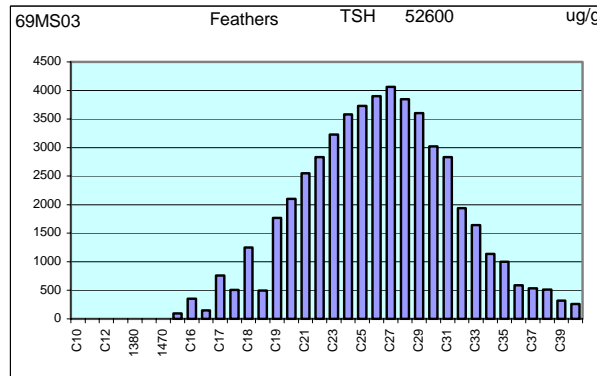
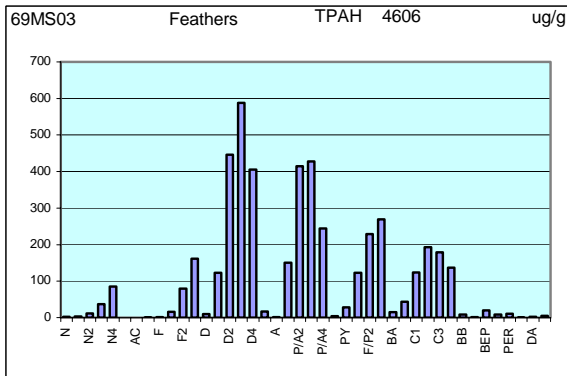
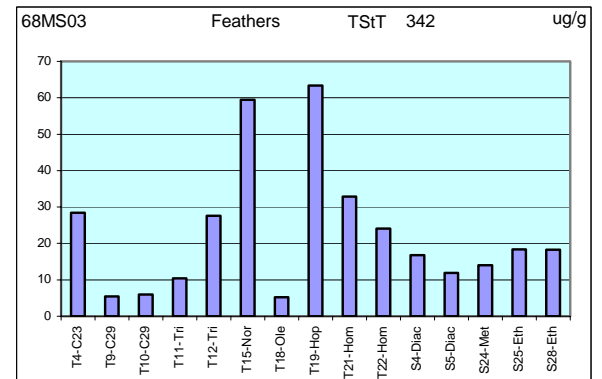
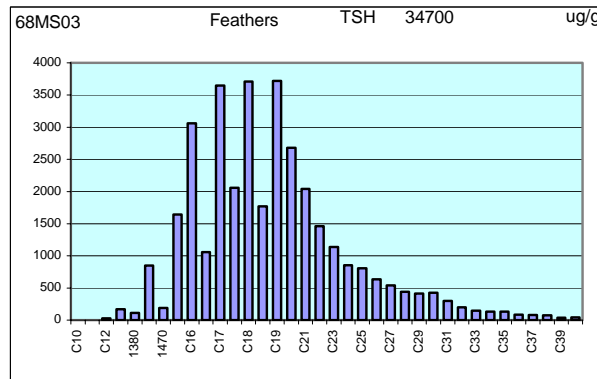
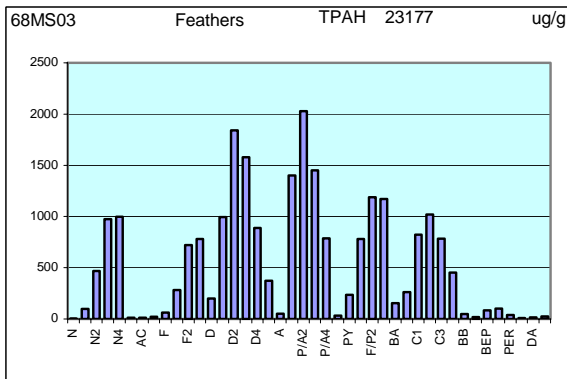
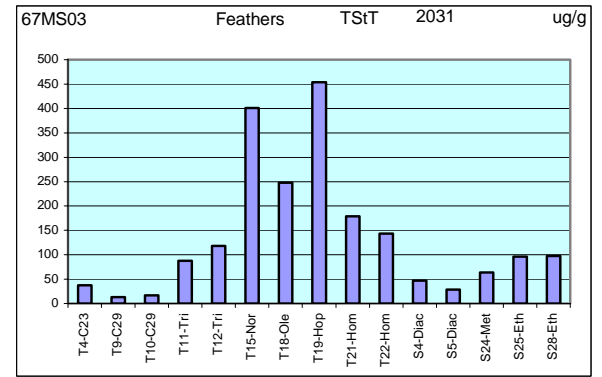
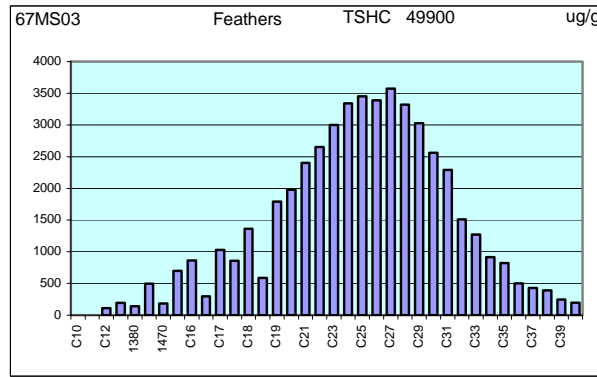
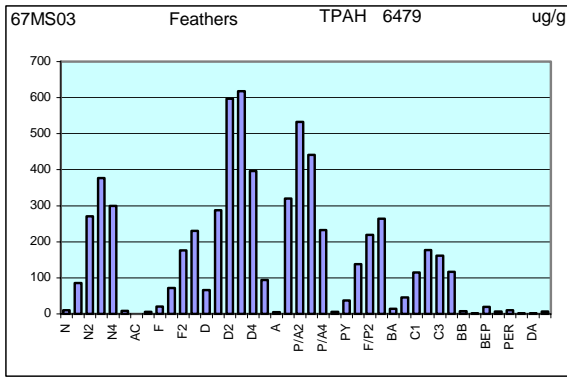


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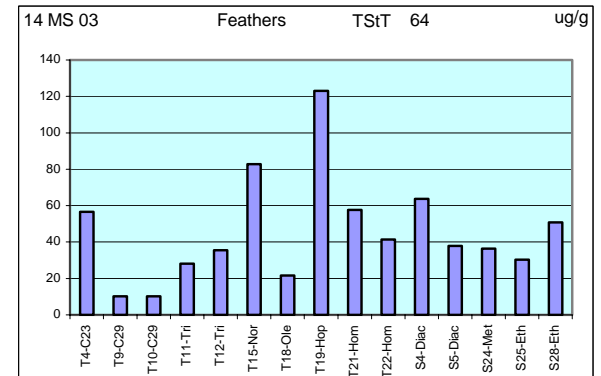
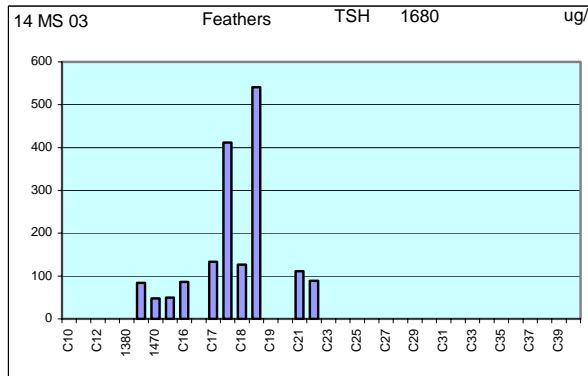
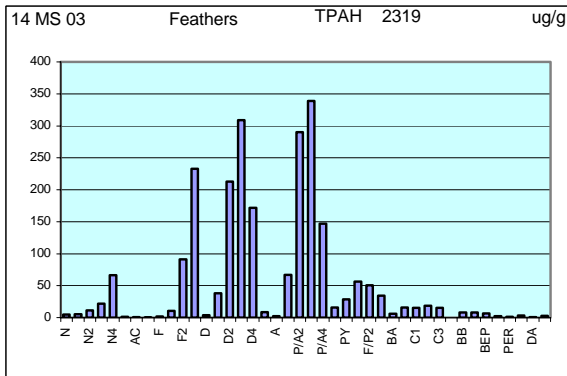
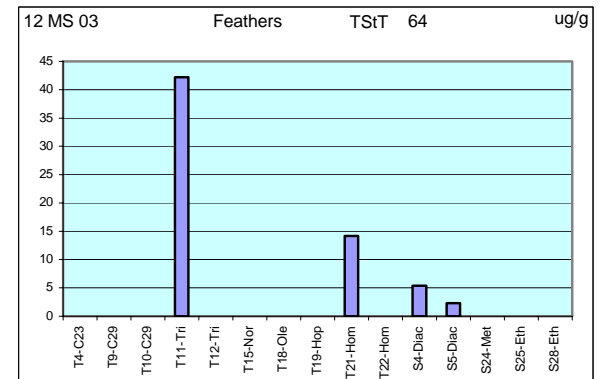
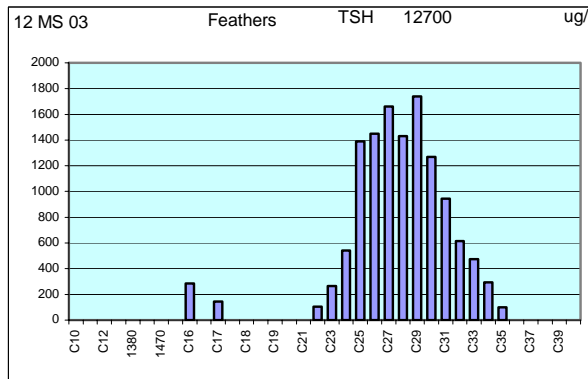
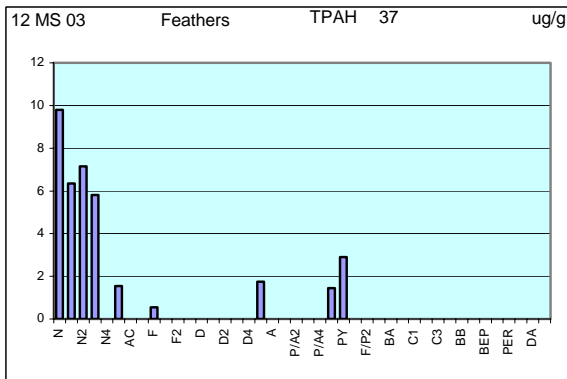
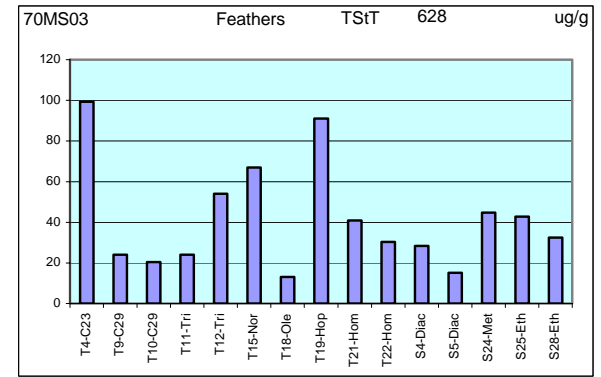
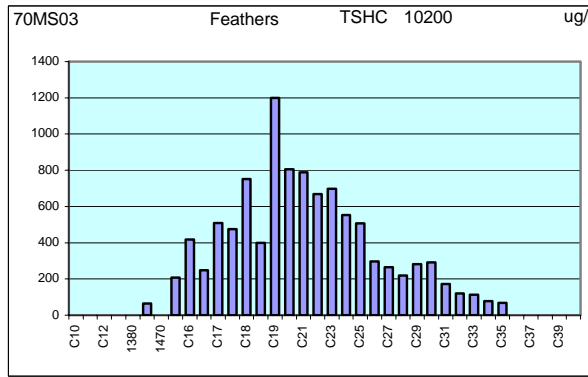
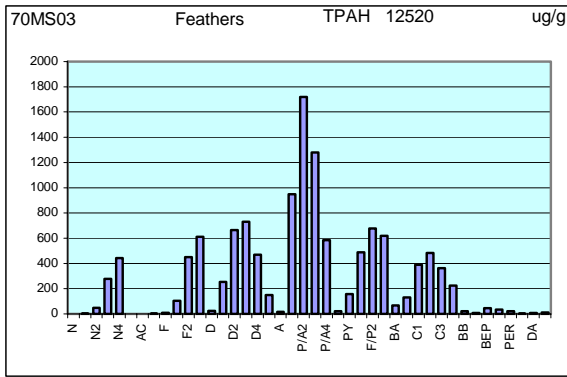




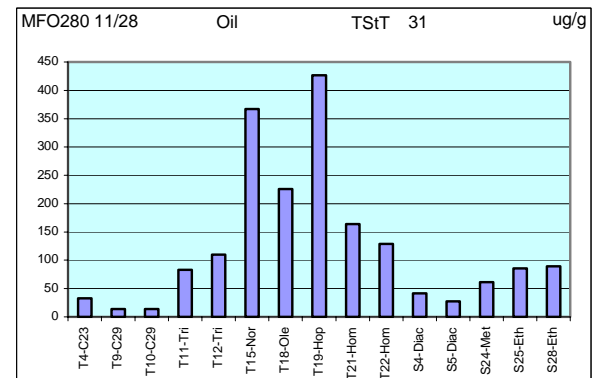
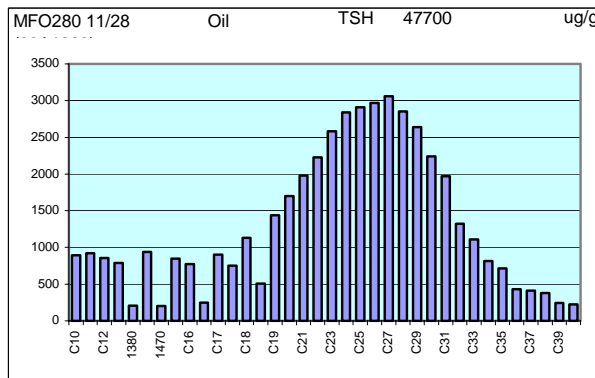
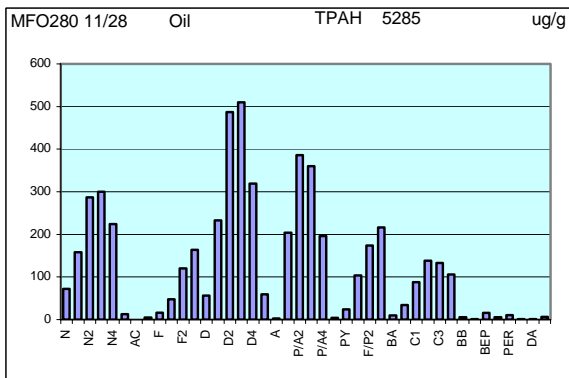
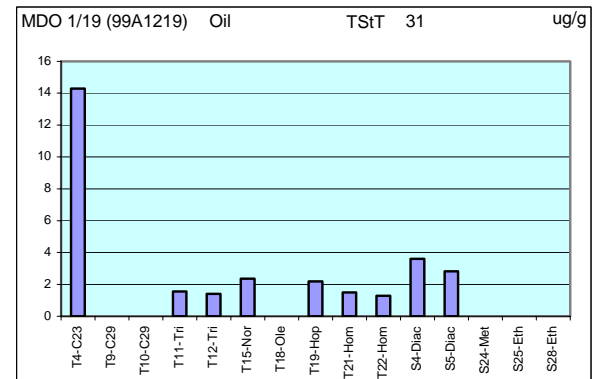
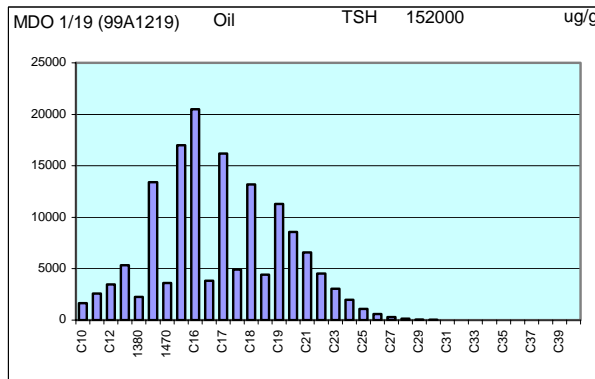
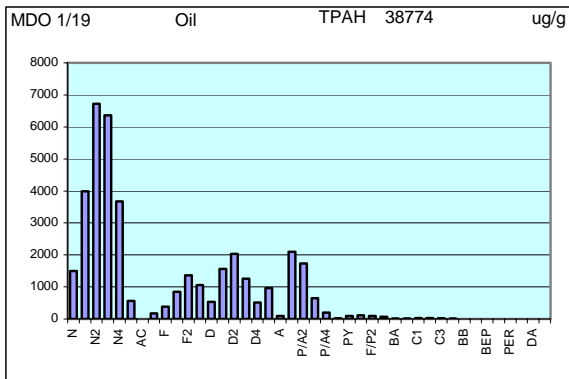
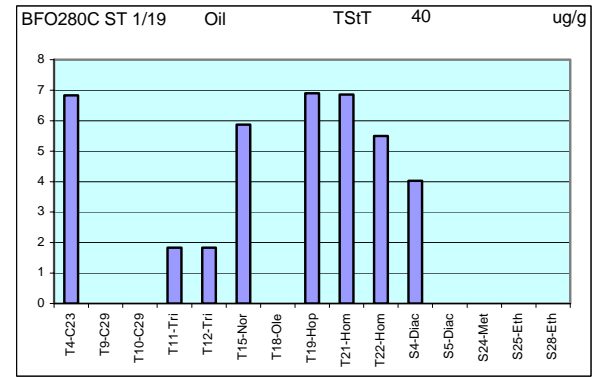
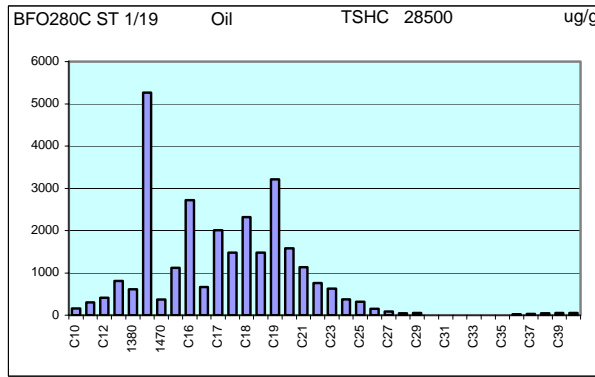
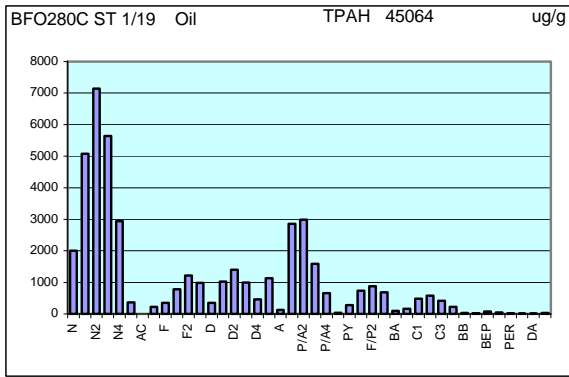
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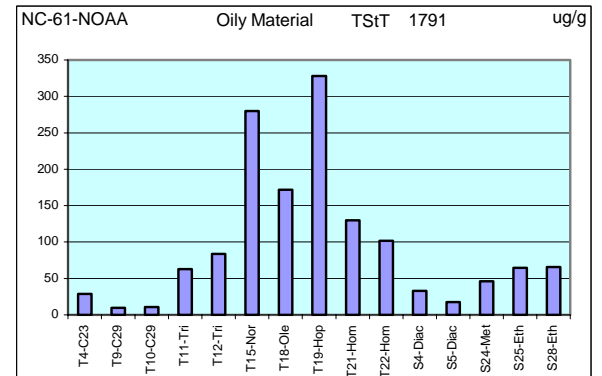
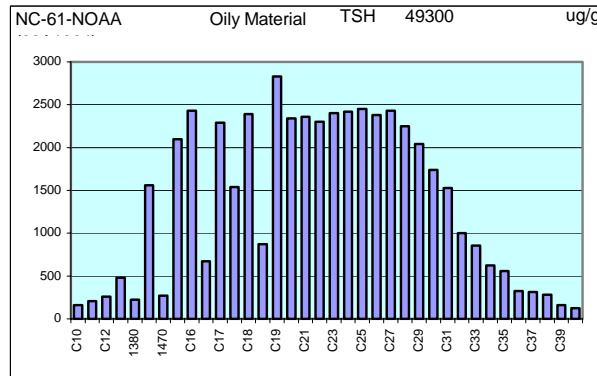
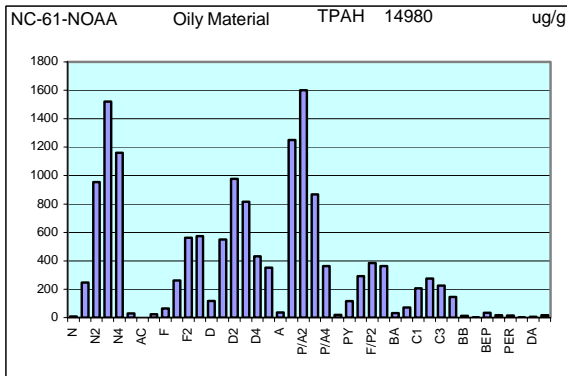
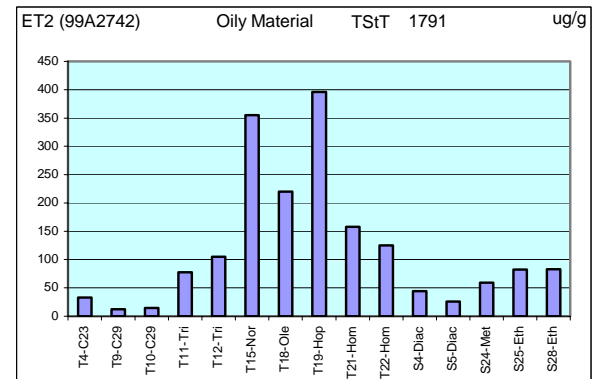
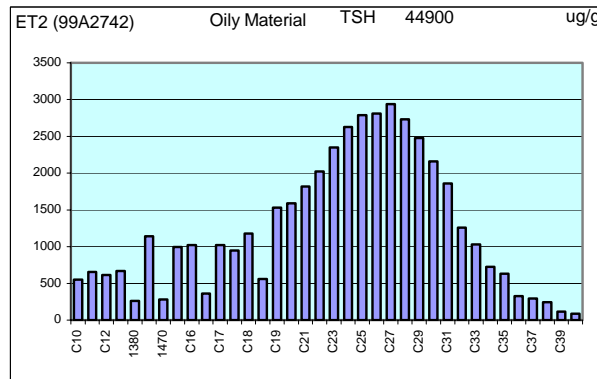
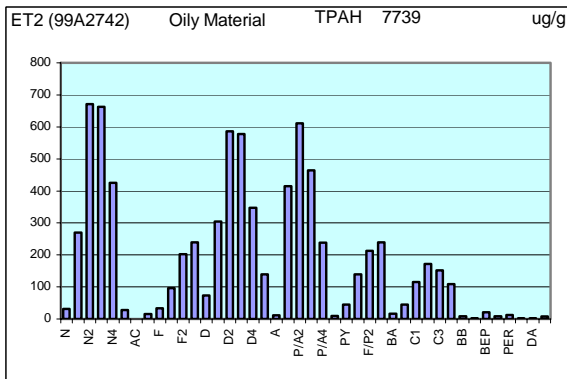
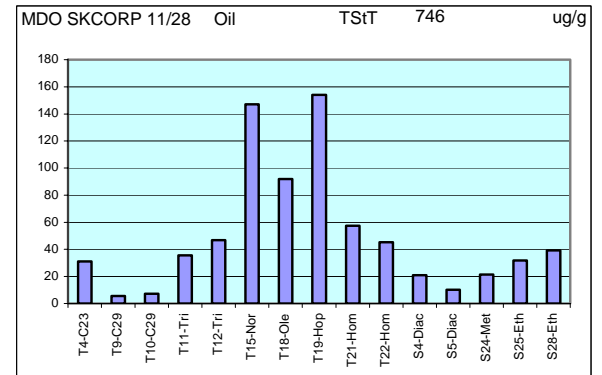
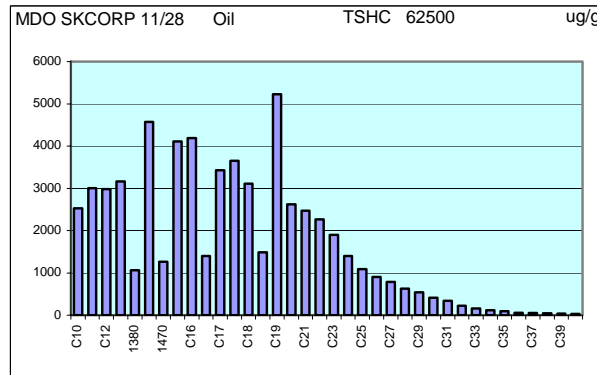
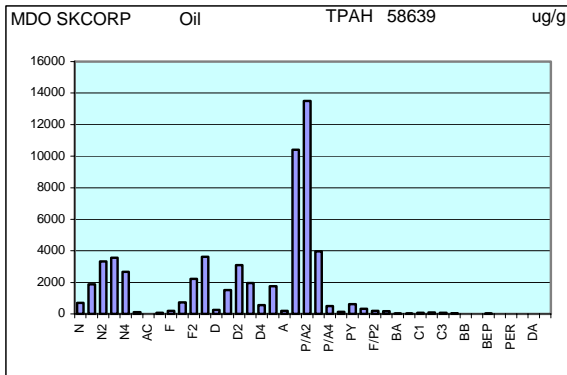
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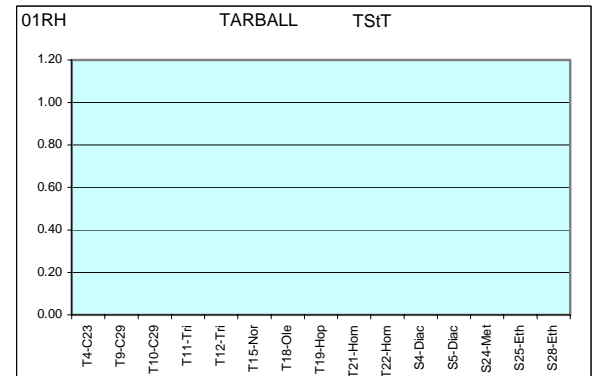
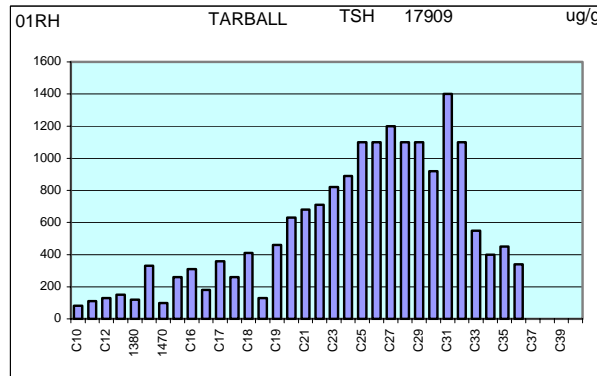
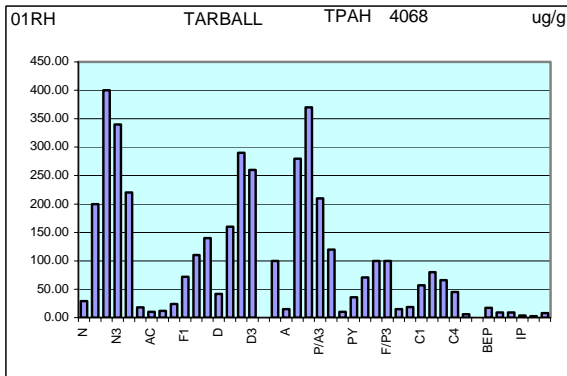
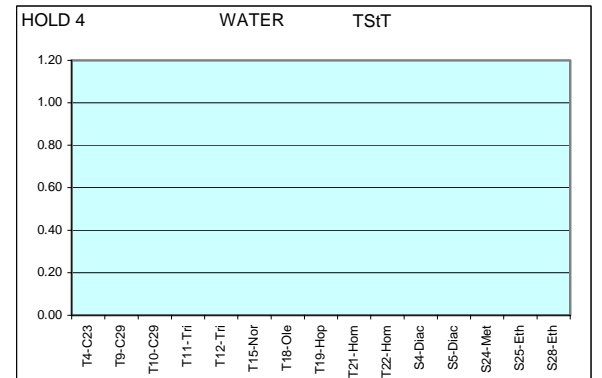
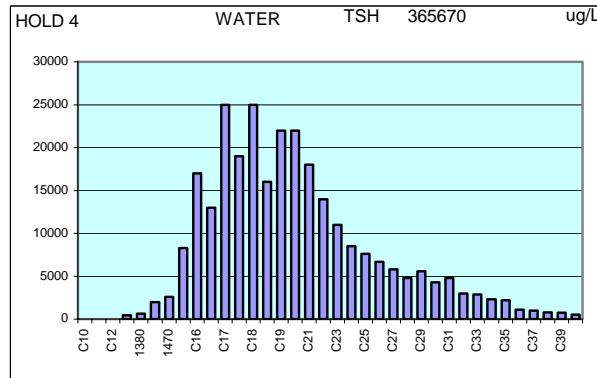
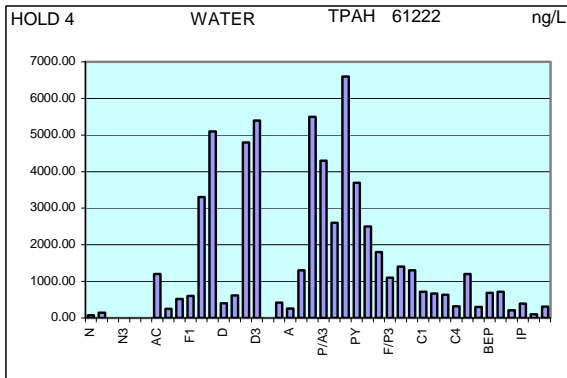
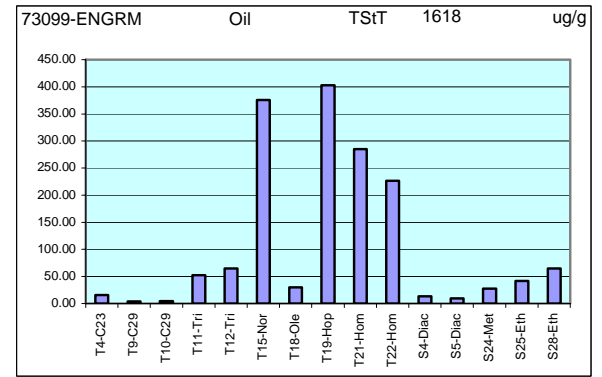
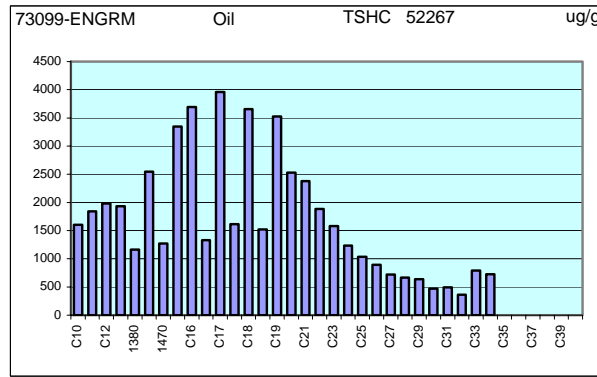
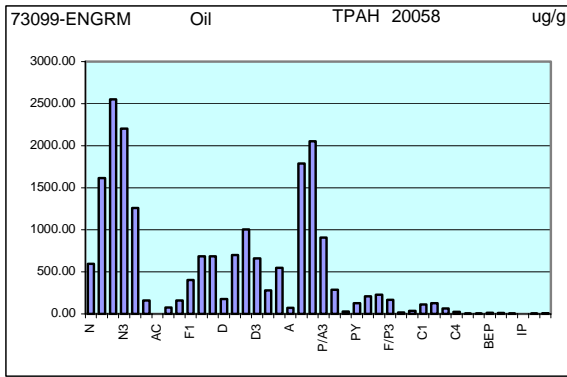
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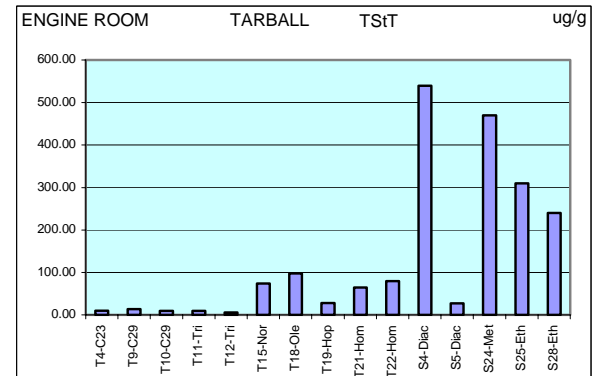
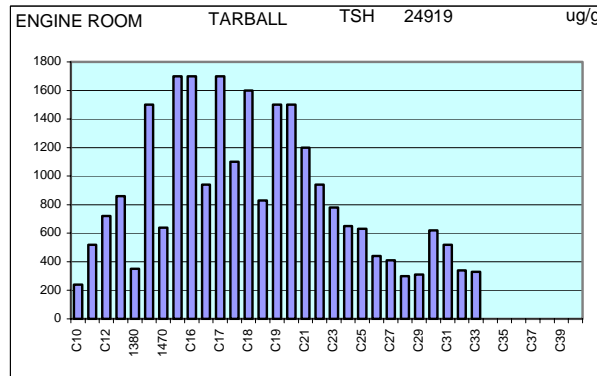
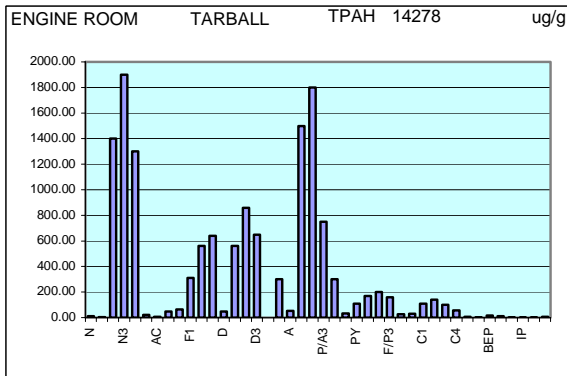
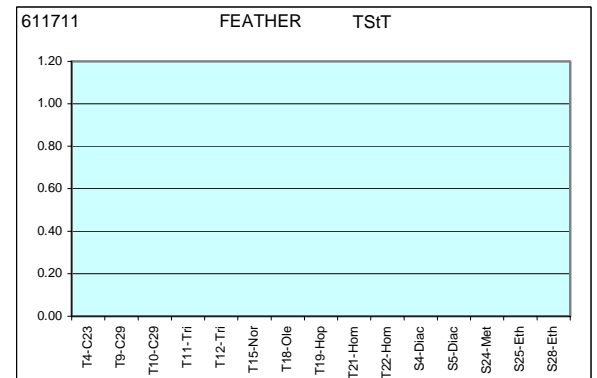
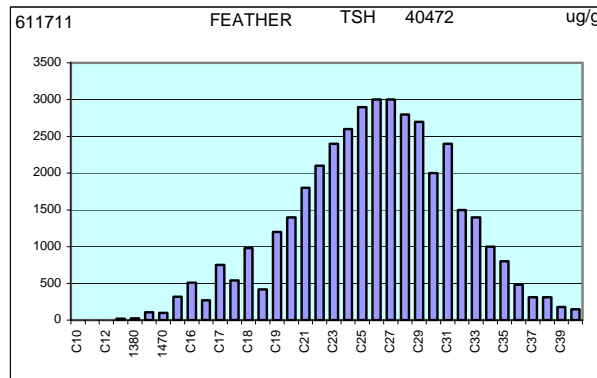
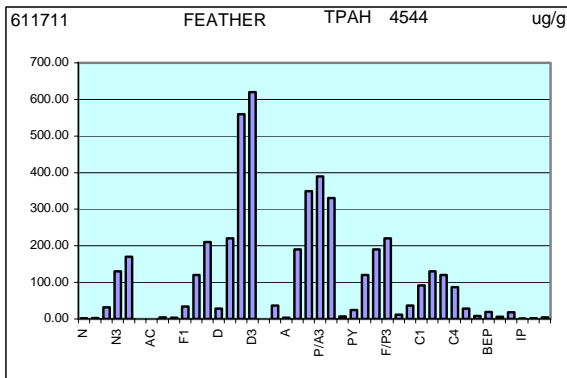
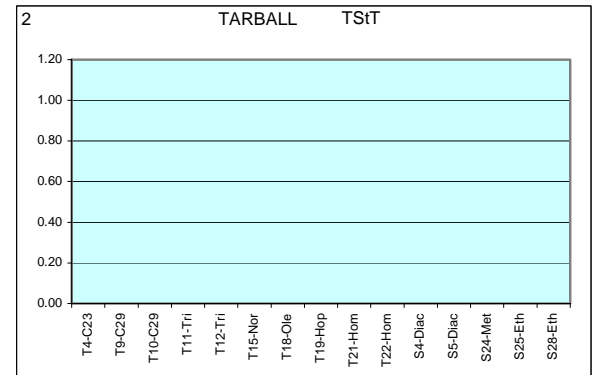
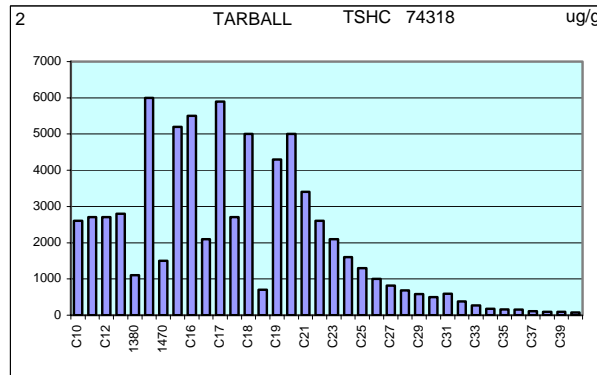
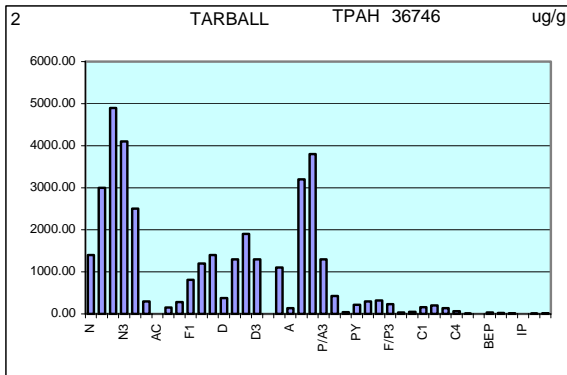
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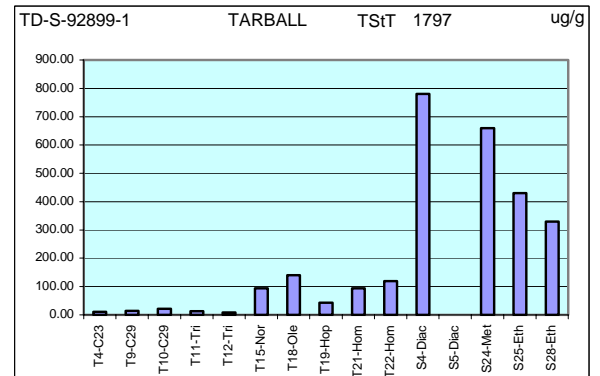
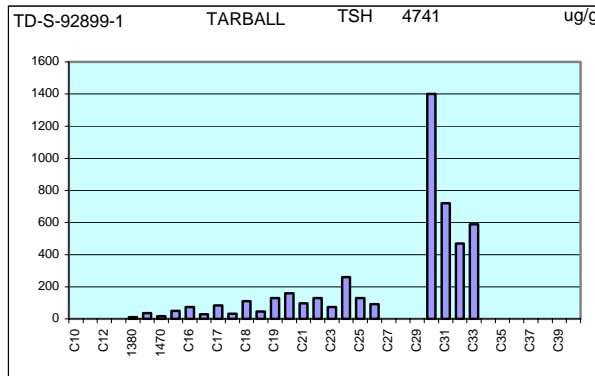
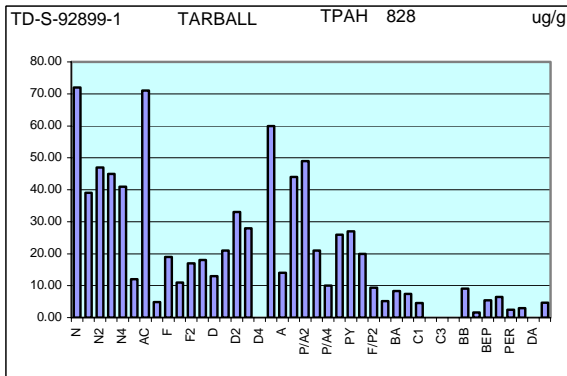
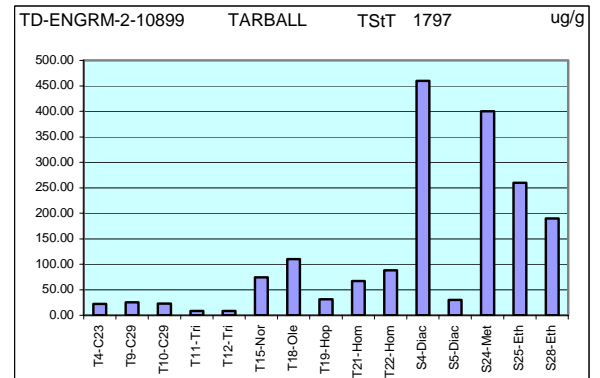
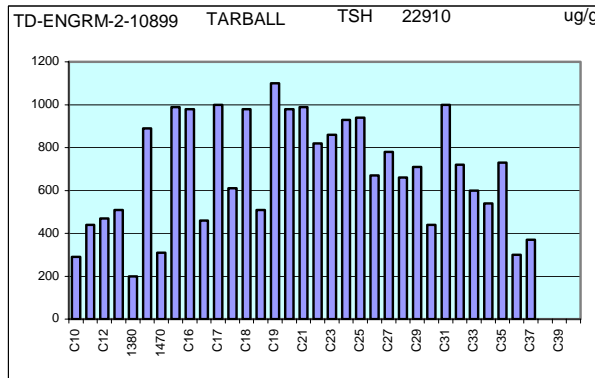
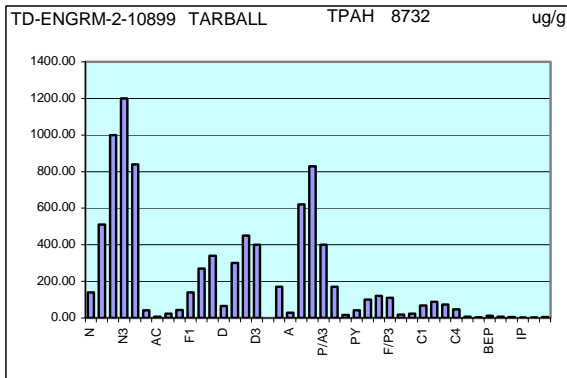
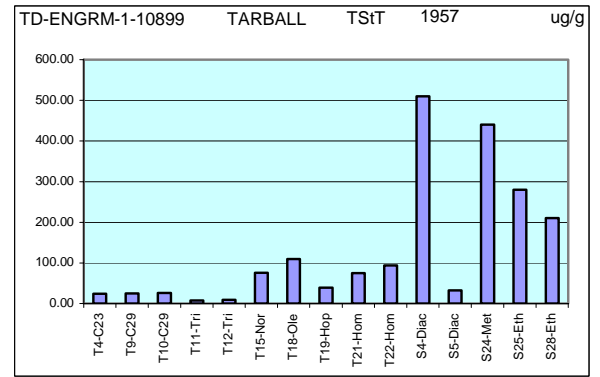
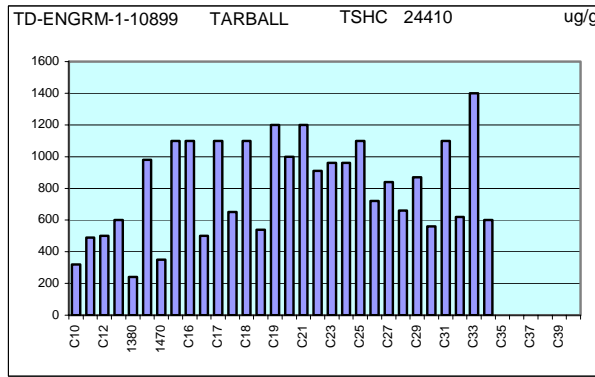
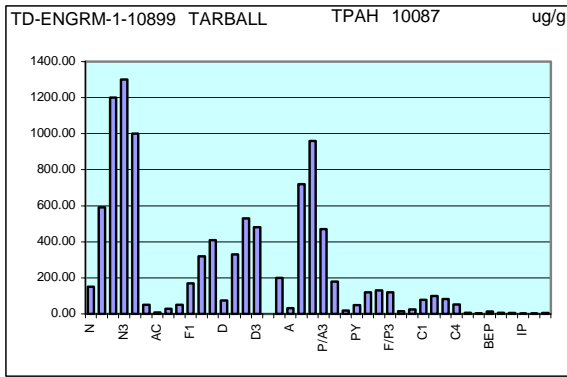
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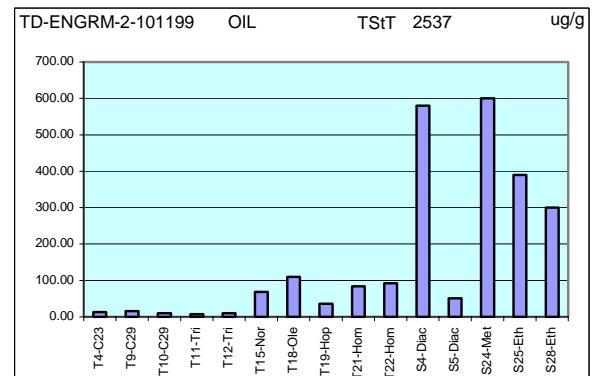
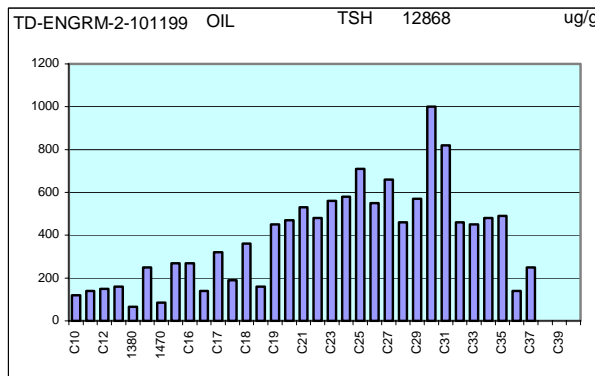
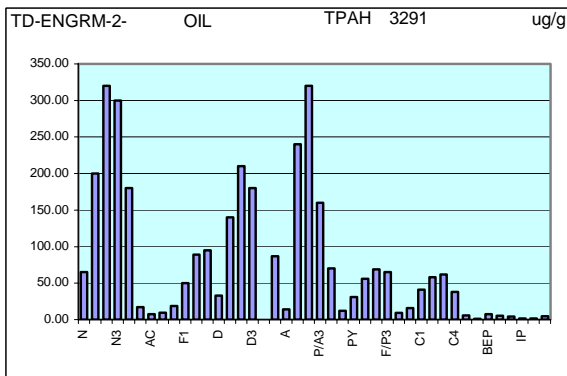
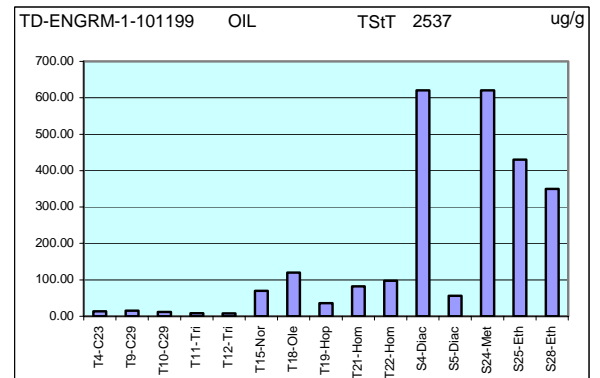
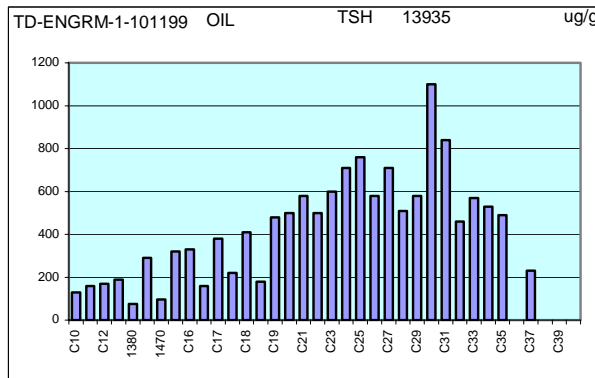
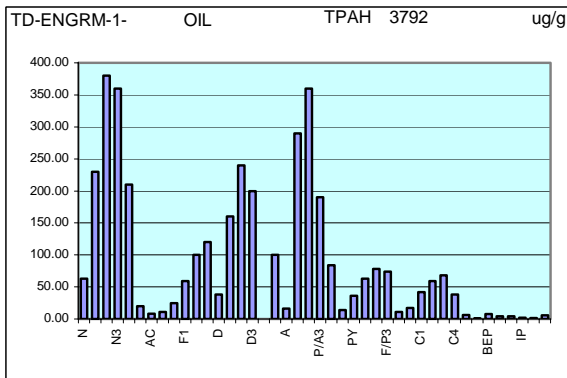
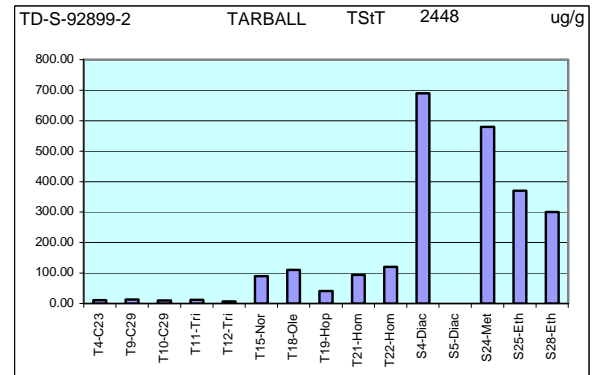
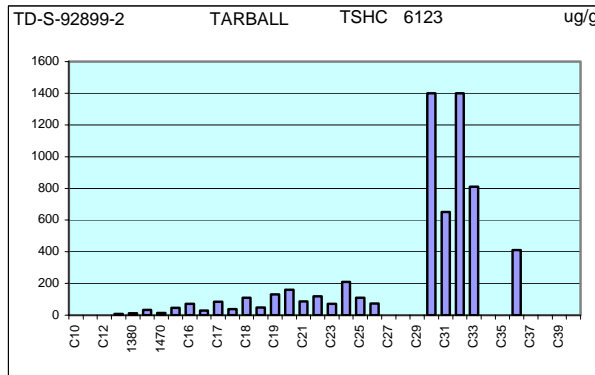
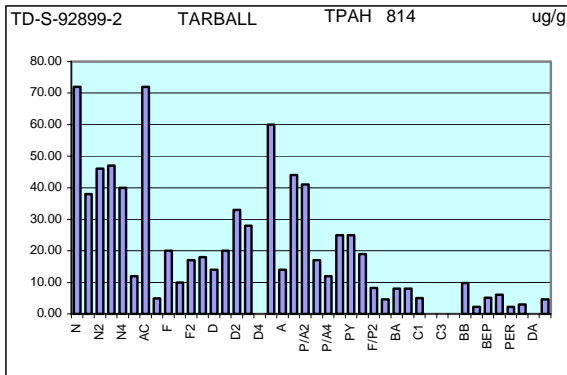
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