

### **5.3 Task 2: Analysis of Plastic Films by ATR-FTIR**

Upon completing task 1 (visual examination and physical measurement), all the heroin substances were transferred into sample bags accordingly whereas the empty case plastic films were analyzed with a HazMatID spectrometer using attenuated total reflectance-Fourier transform infrared spectroscopy (ATR-FTIR). In this task, IR analysis is divided into three sections: 1) qualitative classification of case plastic receptacles based on polymer compositions, 2) instrumental and statistical validation for spectral variation analysis, and 3) statistical spectral variation analysis and classification of the most frequently encountered case plastic type for film-to-film comparison at the distribution/street level.

#### **5.3.1 Qualitative Classification of Case Plastic Films Based on Polymer Compositions**

IR is a relatively useful technique in film analysis because it is insensitive to subtle changes. Therefore, general comparison of films can be meaningfully undertaken despite the subtle distortions (Gilbert *et al.*, 1991) as they do not affect the spectral results. However, the conventional procedure that involves the preparation of films in potassium bromide (KBr) often leads to poor repeatability in IR analysis especially when the amount of film prepared at different times are varied. With FTIR which operates in the ATR mode, errors arising from the preparation of the KBr disc can be greatly minimized. In this technique, the fixed diamond surface that constantly scans the fixed sizes of film samples on a stage helps enhance the precision of the readings.

In this study, each plastic film required less than 2 min to obtain its spectra. Five distinctive types of plastic polymer were identified from the 311 heroin cases, namely Types 1 to 5 (Figure 5.16 and Table 5.4), based on their absorption bands. The true identities of these polymer types were not of interest since the objective of this task was

to discriminate the plastic films rather than to identify them. With reference to the work of Roux *et al.* (2000), absorption bands over the range of 2950 – 2800  $\text{cm}^{-1}$ , 1500 – 1350  $\text{cm}^{-1}$  and 1200 – 800  $\text{cm}^{-1}$  of Types 1 and 5 show the characteristics of a polypropylene backbone. Types 2, 3 and 4 also collectively display the same absorption patterns over the IR range with bands at 2950 – 2800  $\text{cm}^{-1}$ , 1460  $\text{cm}^{-1}$  and 720  $\text{cm}^{-1}$ . These bands suggest a polyethylene backbone. Only one case involved Type 4. As the duplicate IR spectra obtained from this case clearly showed extra absorption bands at 1000 – 1500  $\text{cm}^{-1}$  on the Type 2 backbone, hence it was decided to classify it as a separate type.

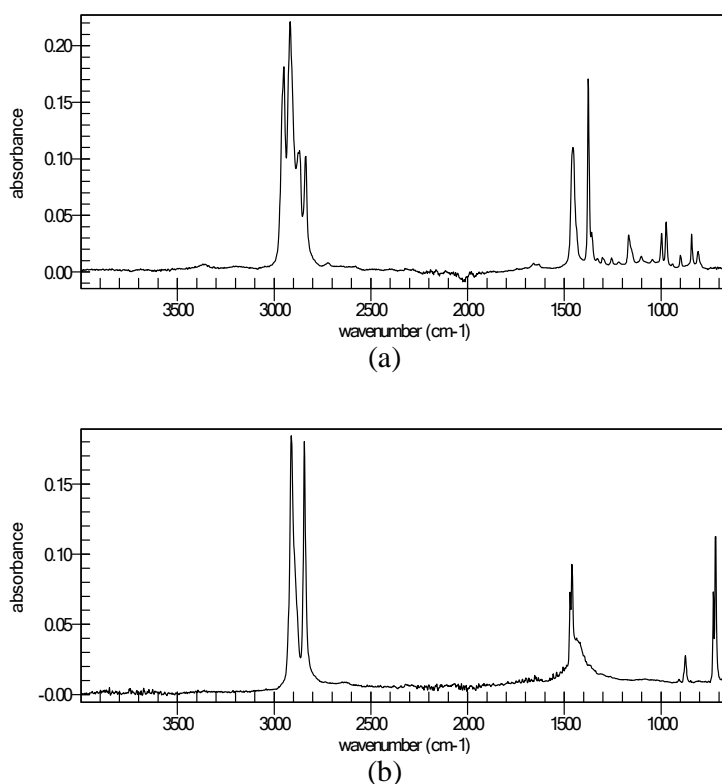


Figure 5.16: Five types of polymeric films encountered in 311 heroin cases (Spectra in (a)-(e) represent Types 1 to 5; Types 1 and 5 are polypropylene-based whereas Types 2 to 4 are polyethylene-based)

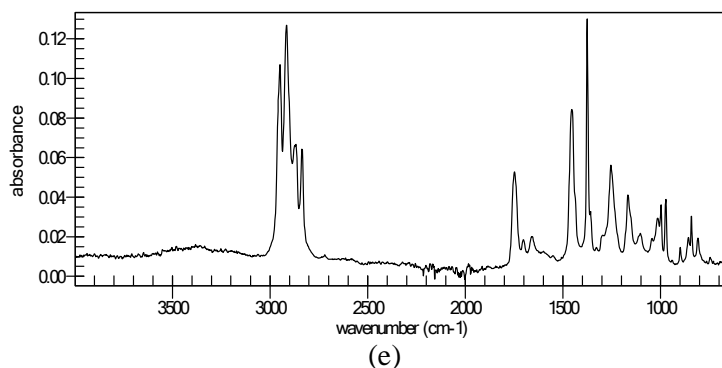
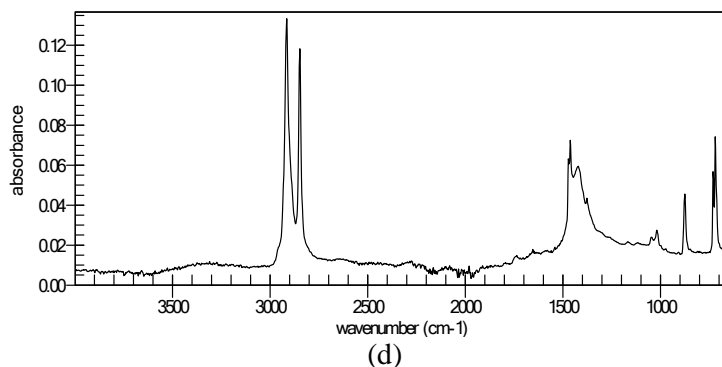
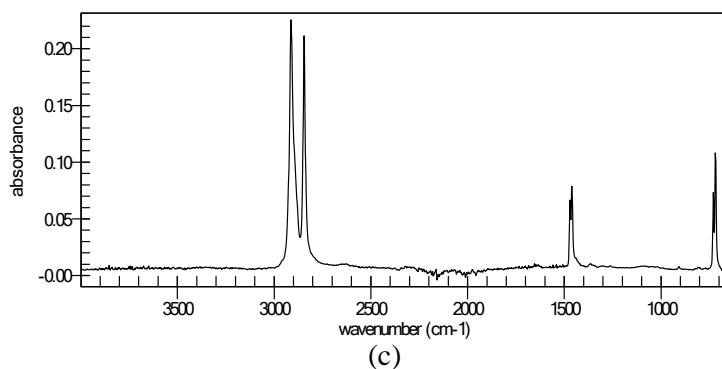


Figure 5.16: Continued

Table 5.4: Polymer types distinguished by ATR-FTIR

Plastic type	Significant peaks in spectrum ( $\text{cm}^{-1}$ )
1	2948, 2914, 2873, 2834, 1456, 1374, 1165, 996, 972, 898, 839, 809
2	2912, 2846, 1460, 873, 718
3	2915, 2848, 1460, 721
4	2915, 2848, 1464, 1422, 1019, 872, 720
5	2948, 2915, 2868, 2837, 1748, 1701, 1660, 1453, 1377, 1254, 1165, 1105, 1016, 997, 972, 895, 856, 842, 809

In some plastic films, weak variations in the spectra of similar polymer type (especially Type 1) were also detected. Causin *et al.* (2006) postulated that these variations could be ascribed to the presence of fillers, co-monomers and pre-oxidants which are added to the base polymers during manufacturing. As these signals were weak, quantitative assessment was not feasible with these minor variations. In addition to the spectral variation, the intensities of the significant peaks of the similar type were also measurably varied and this is addressed in Section 5.3.2 Instrumental and Statistical Validation for Spectral Variation Analysis.

At the distribution level, the plastic types are useful to infer the mode of operation of the heroin packers/distributors in packing the drug. Out of the 311 cases, Type 1 accounted for 92.9% of the cases. However, Type 2 (1.0%), Type 3 (5.1%), Type 4 (0.3%) and Type 5 (0.6%) were relatively less prevalent. The frequency of their encounters in the four geographical regions (Kuala Lumpur = KL, Selangor = SL, Negeri Sembilan = NS and Pahang = PH) are presented in Figure 5.17.

Type 1 constituted the major proportion of the cases submitted by the enforcement units of the four regions. The preference of the heroin packers toward this particular film type could be attributed to the commercial availability of this plastic type at any outlet selling plastic materials and its reasonable strength in holding coarse heroin substances as this film is relatively robust. Besides, the prevalence is also suggestive of cross distribution of heroin packed by a single packer whose *modus operandi* is associated with Type 1 plastic packets. Type 2 was common in KL and NS. Type 3 was the second largest type, although the inclination toward its use remained somewhat inconspicuous. The Type 3 plastic was found in KL, SL and NS but not in PH. The choice of Types 2 and 3 for packing the heroin substance can be ascribed to their universal use among the local hawkers to pack food and hence its ready availability. Types 4 and 5 were rarely chosen, and their occurrence was often

associated with large plastic bags (> 9 cm in width). Both Types 4 and 5 were only found in samples seized in SL and KL respectively. Type 4 was the common type of fragile plastic bags used at supermarkets for packing wet materials, and it is not readily obtainable at every outlet selling plastic materials. Its rare occurrence is due to the reason that this film is thin and it is not adequate for handling heavy masses like heroin substances especially for an undetermined period of distribution. So it is less commonly used for trafficking purposes. The rare occurrence of Type 4 also suggests that this packer was operating on an *ad hoc* basis or on a small scale as the use of this plastic bag had not been evident in other cases/seizures as well as in other regions. Of all the cases, small plastic packets of Type 1 were prominent, and they were selected for spectral variation analysis.

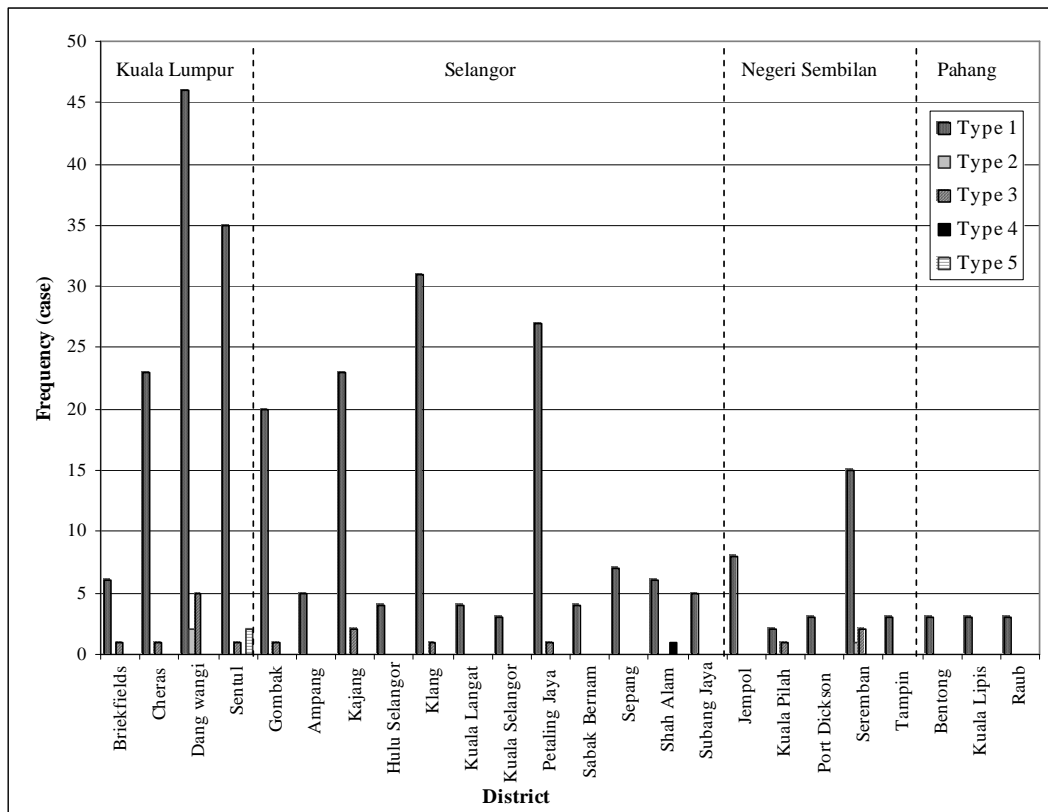


Figure 5.17: Frequency of plastic encounters according to the geographical region in Malaysia

### 5.3.2 Instrumental and Statistical Validation for Spectral Variation Analysis

As previously stated, spectral variations were observed in the Type 1 polypropylene-based films. In line with the concept that IR is less sensitive to changes imparted by environmental influences (Gilburt *et al.*, 1991), so the detected variations could be the result of film compositional variations. Regardless of whether the differences are statistically significant, the instrumental performance and IR data-processing method must be subjected to validation. Data normalization was used to minimize variations imparted by the instrument and environmental factors in the similar films. PCA is an ideal tool for decomposing a large dataset into small but variability-rich dimensions. Therefore, this statistical tool was chosen to maximize the spectral variations displayed by the case plastic films and to evaluate the significant difference between the films. In this sub-task, only the Type 1 polypropylene-based small plastic packets (< 9 cm in width) were considered for linking the films as they are the most recorded in the database. Plastic films of other types were insufficient for PCA.

To ensure true homogeneity of the case films in each case, the variation in the width of the plastic packet in each chosen case should not exceed 0.5 mm. This maximum permitted variation (MPV) is the error associated with the ruler used, and thus homogeneous samples should not show errors greater than this value. This limit also takes into account errors arising from shrinkage or expansion of the films due to environmental influences. However, the length of the plastic packet was not considered as one of the criteria because this variable is often caused by unskilled sealing and is irrelevant to the sample relationship. With this prerequisite, the following steps are proposed for instrumental and statistical validation through IR data pretreatment for spectral data analysis.

### 5.3.2.1 Data Normalization for Minimization of Instrumental and Environmental Influences

Validation for data processing was accomplished using specimen marked 'A' (the 'Spot 1' and 'Spot 2' at different positions). A major issue in the ATR-FTIR data is the differences in the intensities of absorbance caused by optical contact. Figure 5.18 illustrates various effects observed on the spectral outcomes obtained with plastic film A analyzed by ATR-FTIR under five different conditions (four degrees of force, 1X, 2X, 3X, machine press and contamination). Notably, when less force was applied, air gaps created between the plastic film and the diamond surface would lead to less IR-film contact, and thus resulting in lower intensities. The intensities of the peaks increased with increasing force applied. But, they increased as a group rather than as a whole. Four peaks spanning across the range  $2950 - 2800 \text{ cm}^{-1}$  increased in the same proportion while two other peaks in the range  $1500 - 1350 \text{ cm}^{-1}$  increased in another proportion. Therefore, by only taking the signal-to-noise readings without correction could lead to data misinterpretation since the IR data are influenced by poor optical contact arising from uneven film contact on the sample stage. To minimize these errors, data normalization can significantly reduce discrepancies in the peak intensity associated with poor optical contact but less likely with contamination. Besides, Figure 5.18(e) shows that contamination can disrupt the general spectra and affect the small peaks to a greater extent. So three peaks at approximately  $2948 \text{ cm}^{-1}$ ,  $2914 \text{ cm}^{-1}$  and  $2873 \text{ cm}^{-1}$  were normalized with reference to the fourth peak at  $2834 \text{ cm}^{-1}$  (these four signals usually increase/decrease as a group), while the sixth peak at  $1374 \text{ cm}^{-1}$  was normalized to the fifth peak at  $1456 \text{ cm}^{-1}$  (these two signals behave as another group), and all the other small peaks were disregarded due to the relatively high errors associated with the low absorbance intensities. During normalization, the absorbance maxima of the selected peaks processed by the HazMatID software were utilized.

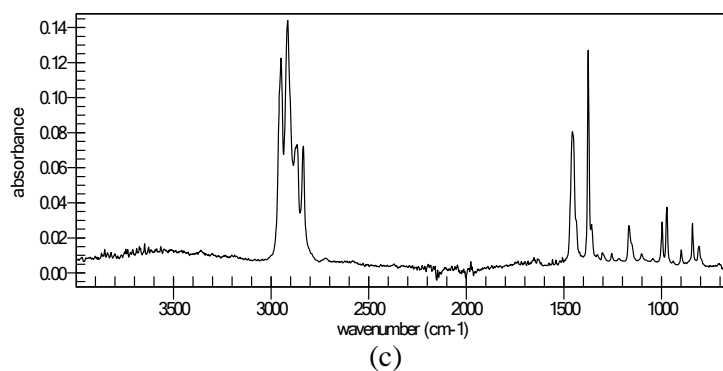
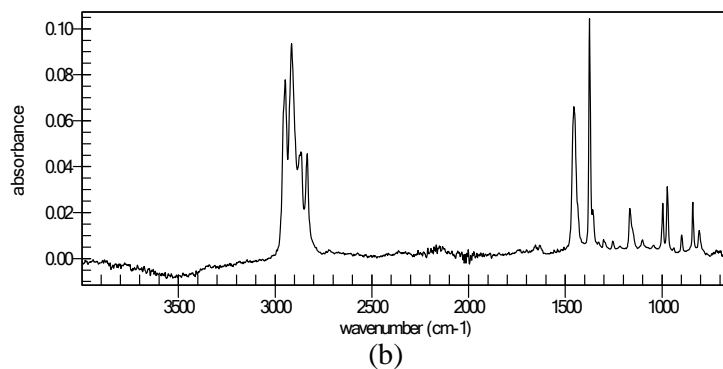
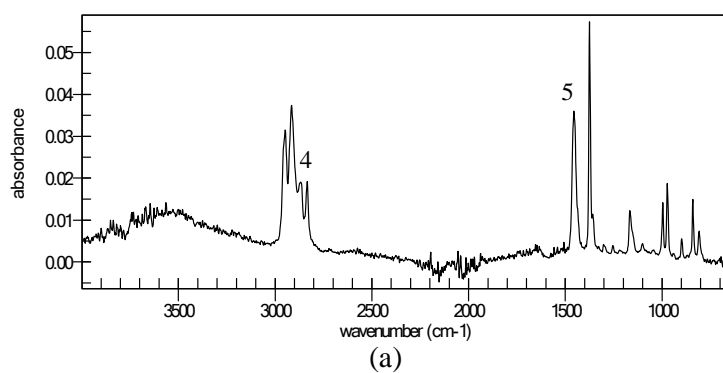
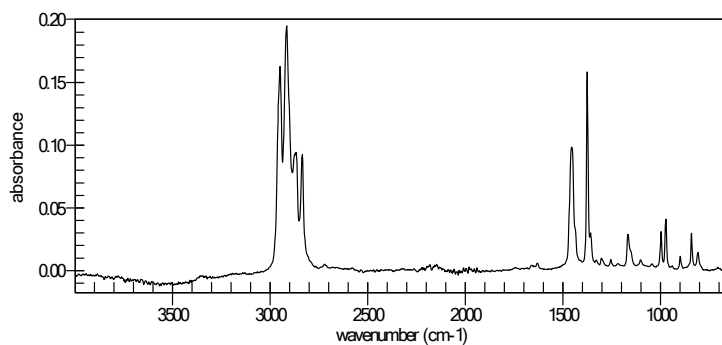
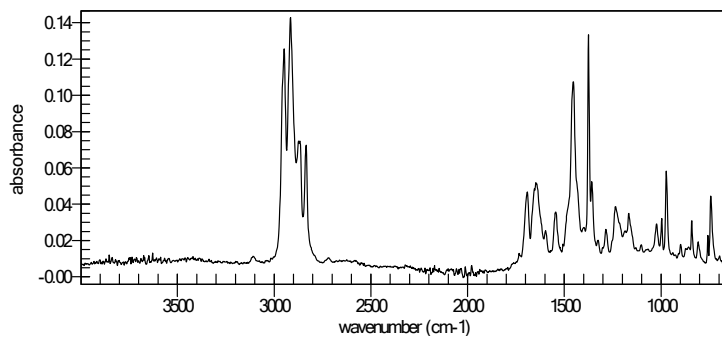


Figure 5.18: Five spectra of Spot 1 obtained under different conditions (Spectra were obtained under (a) 1 X force, (b) 2 X force, (c) 3 X force, (d) machine press and (e) contaminated film; (f) spectra of a street heroin sample highly cut with caffeine which was the characteristic of the local samples; contamination was largely ascribed to the peaks arising from caffeine)

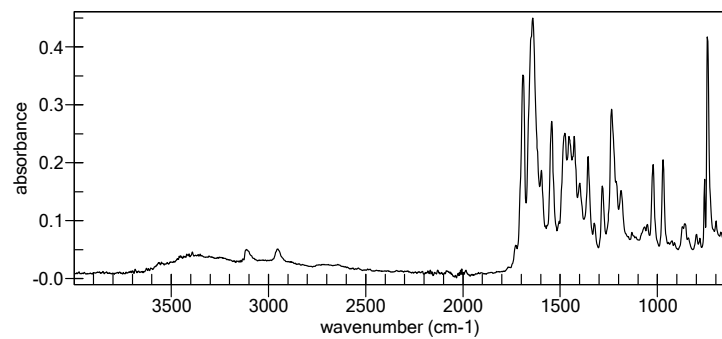




(d)



(e)



(f)

Figure 5.18: Continued

Prior to normalization, Table 5.5 shows that the original intensity of the peak varied significantly when different degrees of force and contamination were applied on the same spot. With the same degree of force, variation was also apparent in Spots 1 and 2 despite both spots originating from the same film. After normalization, all the affected peak intensities of Spots 1 and 2 obtained under varying degrees of force showed a close agreement in the normalized form (Table 5.5), except especially the sixth peak which was affected by contamination. Therefore, data normalization is particularly

useful to minimize the effects of uneven force or poor optical contact. As the diamond surface and plastic films were thoroughly cleaned prior to being scanned, hence contaminated spectra were not a major concern in this study. In addition, the contamination-free spectra and result consistency shown by two representative spectra obtained from each case have confirmed the absence of contamination during analysis.

Table 5.5: Normalized data and original peak intensities in parentheses of four selected peaks obtained from Spots 1 and 2 of specimen marked ‘A’

Spot	1				2			
	Original Peak number	1	2	3	6	1	2	3
<b>1 X Force</b>	1.68 (0.032)	1.95 (0.037)	1.00 (0.019)	1.58 (0.057)	1.70 (0.034)	2.05 (0.041)	1.05 (0.021)	1.61 (0.058)
<b>2 X Force</b>	1.70 (0.078)	2.04 (0.094)	1.00 (0.046)	1.58 (0.104)	1.67 (0.050)	2.03 (0.061)	1.00 (0.030)	1.60 (0.075)
<b>3 X Force</b>	1.69 (0.122)	2.00 (0.144)	1.01 (0.073)	1.59 (0.127)	1.71 (0.087)	2.14 (0.109)	1.00 (0.051)	1.61 (0.113)
<b>Machine press (MP)</b>	1.75 (0.163)	2.10 (0.195)	1.00 (0.093)	1.62 (0.159)	1.73 (0.182)	2.15 (0.226)	1.02 (0.107)	1.59 (0.173)
<b>Contaminated</b>	1.73 (0.126)	1.96 (0.143)	1.03 (0.075)	1.24 (0.133)	1.76 (0.123)	1.93 (0.135)	1.04 (0.073)	0.86 (0.143)
<b>% Δ Normalized MP</b>	<sup>a</sup> 1.16%	2.38%	2.00%	1.89%	-	-	-	-
<b>% Δ Original MP</b>	<sup>b</sup> 11.66%	15.90%	15.05%	8.81%	-	-	-	-

<sup>a</sup>Degree of difference between the first normalized peaks of Spots 1 and 2 under machine press:  $[(1.75-1.73)/1.73 \times 100] = 1.16\%$

<sup>b</sup>Degree of difference between the first original peak intensities of Spots 1 and 2 under machine press:  $[(0.182-0.163)/0.163 \times 100] = 11.66\%$

In practice, the machine press was used. When the difference in the normalized peak intensity between two corresponding peaks of Spots 1 and 2 measured under machine press is expressed as a percentage, Table 5.5 reveals that the percentage difference in the normalized data is smaller than the percentage difference in the original peak intensities. This smaller difference is desirable as it represents the minimum intra-batch variation for two related film materials (e.g. Spots 1 and 2 of specimen A). In contrast, it is expected that the normalized data of unrelated plastic

films (from different sources) will show significant differences (inter-batch variation). The signal differences are assumed to be resulted from the difference in the film material rather than from other factors.

### **5.3.2.2 Repeatability and Reproducibility Improved by Data Normalization**

Seven specimens marked 'B' were used to assess the precision of the normalized data. This precision represents the repeatability (intra-day variation) and reproducibility (inter-day variation) of the instrument. It also indicates intra-batch variability as different film specimens originating from a single source were repeatedly measured. The intra-day precision was expressed as the RSD which was calculated from 8 sets of original and normalized data obtained from the seven specimens measured on the same day. For the inter-day precision, the same calculation was also applied to 20 sets of original and normalized data obtained from the seven specimens measured on different days. Table 5.6 shows that the instrument was repeatable ( $RSD < 2\%$ ) irrespective of whether the data were normalized. However, the inter-day precision was poor ( $RSD < 8\%$ ) when the original peak intensities were used. This was improved through normalizing the target peaks. After the normalization, the inter-day precision ( $RSD < 3\%$ ) of the instrument has been improved significantly. This precision also indicates the improvement of intra-batch variation shown by the seven related individual films originating from the same batch. In this subtask, this variation was used to represent the maximum permitted deviation in film analysis.

In summary, data pretreatment using normalization is useful in reducing errors associated with uncontrolled variables (e.g. uneven force) as well as environmental influences on the related films (e.g. exposure to sunlight).

Table 5.6: RSD (%) of original peak intensities and normalized peaks for specimens B

Peak no.		1	2	3	4	5	6
<b>Original peak intensities</b>							
<b>Intra-day precision (n = 8)</b>	<b>Mean ± SD</b>	0.183 ± 0.002	0.220 ± 0.004	0.108 ± 0.002	0.104 ± 0.001	0.110 ± 0.001	0.174 ± 0.002
	<b>RSD (%)</b>	1.07	1.97	1.74	0.80	0.81	1.34
	<b>Mean ± SD</b>	0.175 ± 0.012	0.210 ± 0.016	0.103 ± 0.007	0.100 ± 0.007	0.106 ± 0.005	0.168 ± 0.009
<b>Inter-day precision (n = 20)</b>	<b>RSD (%)</b>	6.96	7.79	6.64	6.65	4.62	5.58
	<b>Normalized peaks</b>						
<b>Intra-day precision (n = 8)</b>	<b>Mean ± SD</b>	1.759 ± 0.012	2.115 ± 0.029	1.038 ± 0.012	-	-	1.585 ± 0.014
	<b>RSD (%)</b>	0.68	1.38	1.19	-	-	0.89
<b>Inter-day precision (n = 20)</b>	<b>Mean ± SD</b>	1.754 ± 0.014	2.111 ± 0.043	1.039 ± 0.012	-	-	1.578 ± 0.024
	<b>RSD (%)</b>	0.80	2.02	1.13	-	-	1.49

### 5.3.2.3 Statistical Evaluation of Normalized Data by PCA

When the data of specimen A are decomposed by PCA, close relationships among the data points should be resulted, despite different force conditions. On the other hand, the data points of specimens B measured under normal condition (machine press) should also be evenly and closely spaced to show natural random variation on the score plot. Figure 5.19 illustrates how normalization enhances the validity of the IR data. Ideally, the normalized data will reduce the risks of false clusters (unwanted separation) within the same group and improve the relationships of the affected data of the same group. Four score plots of two datasets (respectively from the plastic films A and B) accompanied by their respective percentage variability expressed as %V<sub>1</sub> (first component) and %V<sub>2</sub> (second component) are shown in Figure 5.19. The data points of specimens B are represented by black dots. Whereas the data points of specimen A are

expressed as pairs of bright shapes according to the five conditions specified in Table 5.5.

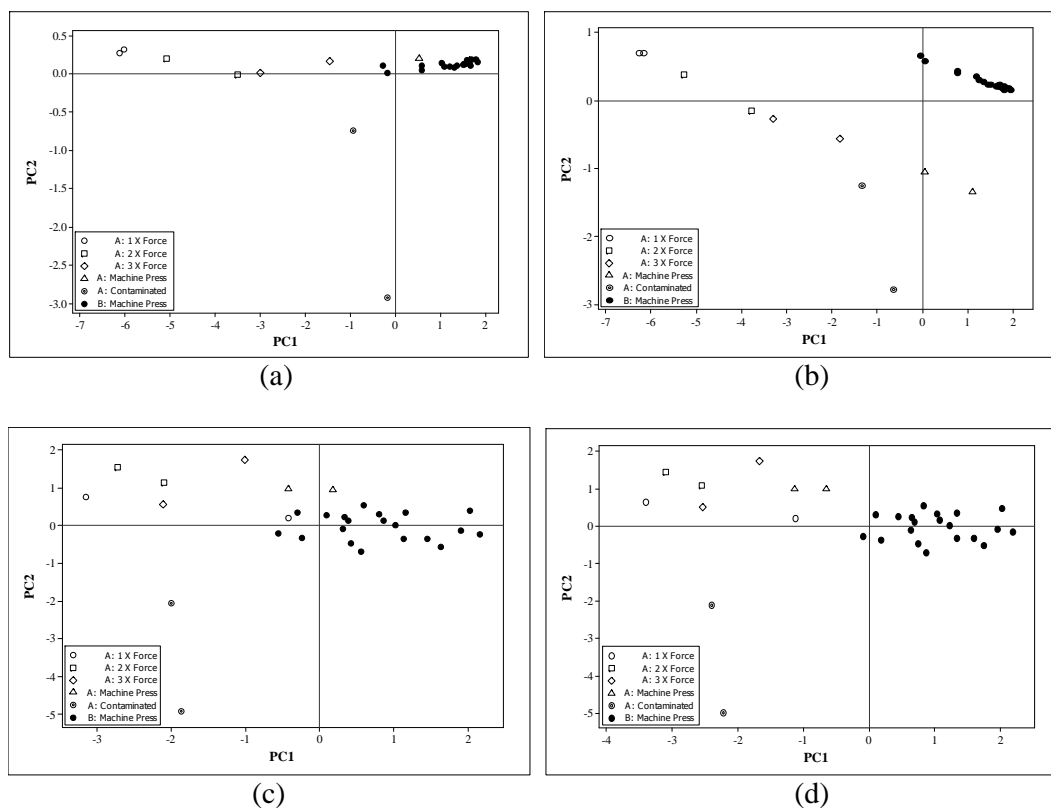


Figure 5.19: Score plots of PCA (correlation mode) decomposed IR data obtained under different conditions (Data distributions vary according to (a) original peak intensities,  $\%V_1 = 94.24\%$  and  $\%V_2 = 5.60\%$ ; (b) original peak intensities and widths,  $\%V_1 = 89.09\%$  and  $\%V_2 = 7.74\%$ ; (c) normalized peak intensities,  $\%V_1 = 50.71\%$  and  $\%V_2 = 34.96\%$ ; (d) normalized peak intensities with widths;  $\%V_1 = 55.08\%$  and  $\%V_2 = 28.00\%$ )

Based on the score plots, the difficulties associated with the original peak intensities can be inferred as follows:

- i. Figures 5.19(a) and 5.19(b) clearly display three clusters of specimens B with the original peak intensities, although only 20 findings were obtained under the machine press condition.

- ii. The pair of specimen A under the machine press condition (“bright triangles”) in Figure 5.19(a) overlaps specimens B. However, they were separated from specimens B when a width variable was added as shown in Figure 5.19(b).
- iii. The score plots in Figures 5.19(a) and 5.19(b) indicate exaggerated long-range relationships for the data points of specimen A that were affected by the force and contamination.

Data normalization can help to solve the following problems:

- i. After data normalization, the false clusters of specimens B were eliminated, and an even distribution was obtained (Figures 5.19(c) and 5.19(d)).
- ii. When all data were measured under the machine press condition, normalized IR data alone or together with the width measurements are sufficient to contribute an ideal spread for the clustering of two distinct groups of data. Bright triangles of normalized specimens A have been separated from specimens B as shown in Figure 5.19(c) and further separated by the width measurement as in Figure 5.19(d). Normalized IR data are therefore an optimized form for clustering similar types of polymeric films and with enhanced discriminative power in the presence of additional variables (e.g. width). Other variables such as film thickness are not discussed because this task was aimed at making use of spectral differences.
- iii. The first component axes in Figures 5.19(c) and 5.19(d) also indicate that the distances between the findings of specimen A are significantly reduced with a

small expansion of the second component axes and hence a closer spread of the data points of specimen A. Normalization is therefore vital to reduce distances arising from any deviations attributed to uncontrolled factors such as uneven force.

Based on the above contention, it is proposed to use this data normalization method for spectral variation analysis. In this subtask, only the first two components of PCA were consistently employed for comparison because the subsequent components may not be meaningful with their negligible variability in certain cases (e.g. score plots in Figures 5.19(a) and 5.19(b) respectively retain a total variability of 99.84% and 96.83% in their first two components).

### **5.3.3 IR Spectral Variation Analysis for the Classification of Case Plastic Films**

The ability of the HazMatID software to retrieve quantitative data further reinforced data analysis. First, 197 cases (having plastic packets within the MPV) were carefully identified. For each case, the mean values of four normalized peak intensities (peaks 1, 2, and 3 were normalized to 4; peak 6 was normalized to 5) of two representative spectra retrieved and processed by the HazMatID software were chosen for statistical reduction. A total of 197 cases multiplied by 4 normalized peaks = 788 variables were directly decomposed by the PCA. Although most normalized data appear to display negligible differences in the selected case, the symbiosis of PCA and IR spectra can detect small spectral differences between these films (Hori & Sugiyama, 2003) and differentiate them through these small differences.

As the inclusion of non-spectral variables such as width will affect spectral variation analysis using the PCA, the spectral differences shown by the case plastic packets were analyzed according to their widths in Figure 5.20.

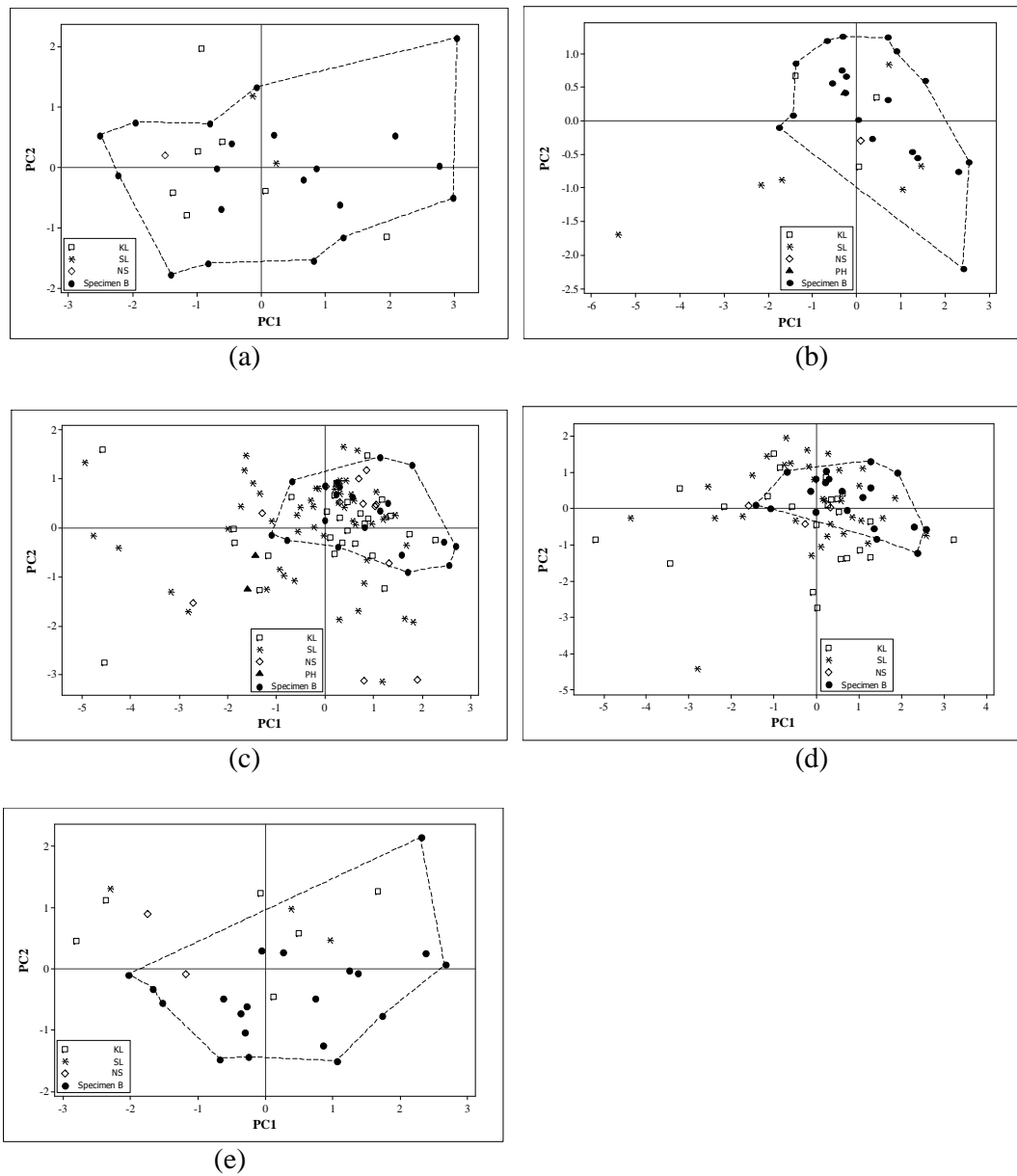
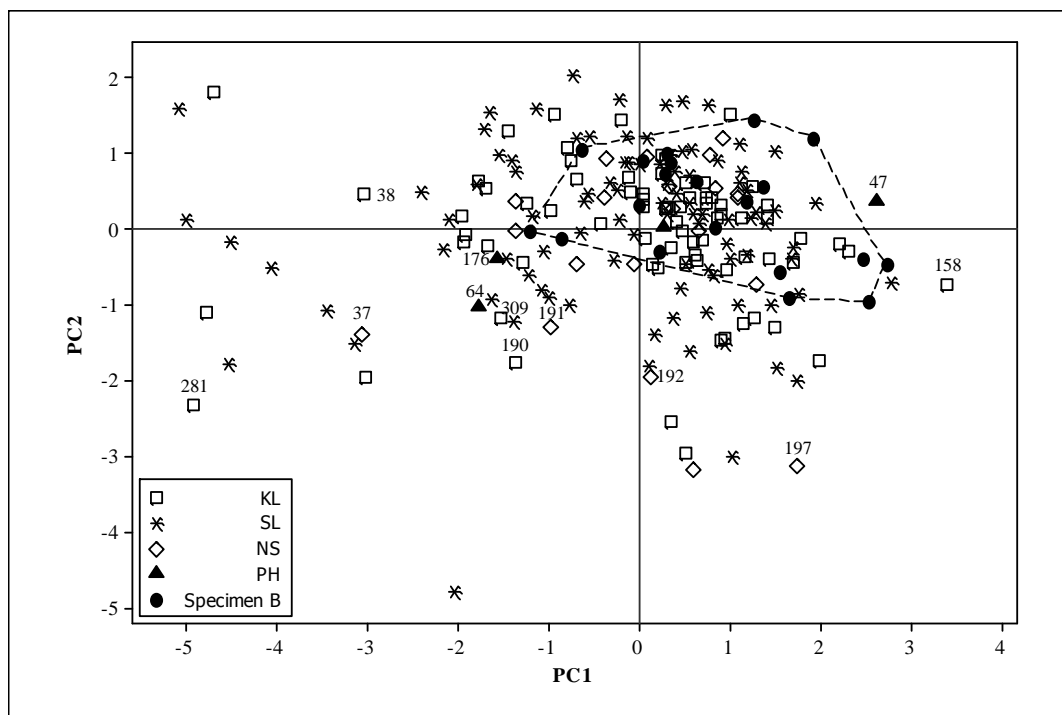


Figure 5.20: Score plots of PCA (correlation mode) decomposed data, the spectral variations measured in Type 1 plastic packets in their respective widths (The score plots respectively represent (a) 10 cases (4.8 – 4.89 cm) with  $\%V_1 = 56.86\%$  and  $\%V_2 = 23.18\%$ ; (b) 11 cases (6.10 – 6.19 cm) with  $\%V_1 = 63.41\%$  and  $\%V_2 = 18.91\%$ ; (c) 92 cases (6.20 – 6.29 cm) with  $\%V_1 = 57.86\%$  and  $\%V_2 = 25.25\%$ ; (d) 58 cases (6.30 – 6.39 cm)  $\%V_1 = 57.65\%$  and  $\%V_2 = 27.56\%$ ; and (e) 11 cases (6.40 – 6.49 cm) with  $\%V_1 = 53.83\%$  and  $\%V_2 = 20.92\%$ ; the widths were decided based on the 197 cases considered; other widths were not included because of their absence in the dataset or low prevalence for PCA; score plots (a) and (e) show natural variation rather than the difference in films when natural variation is defined by the rotated polygon; score plots (c) and (d) have significant differences in certain plastic films)

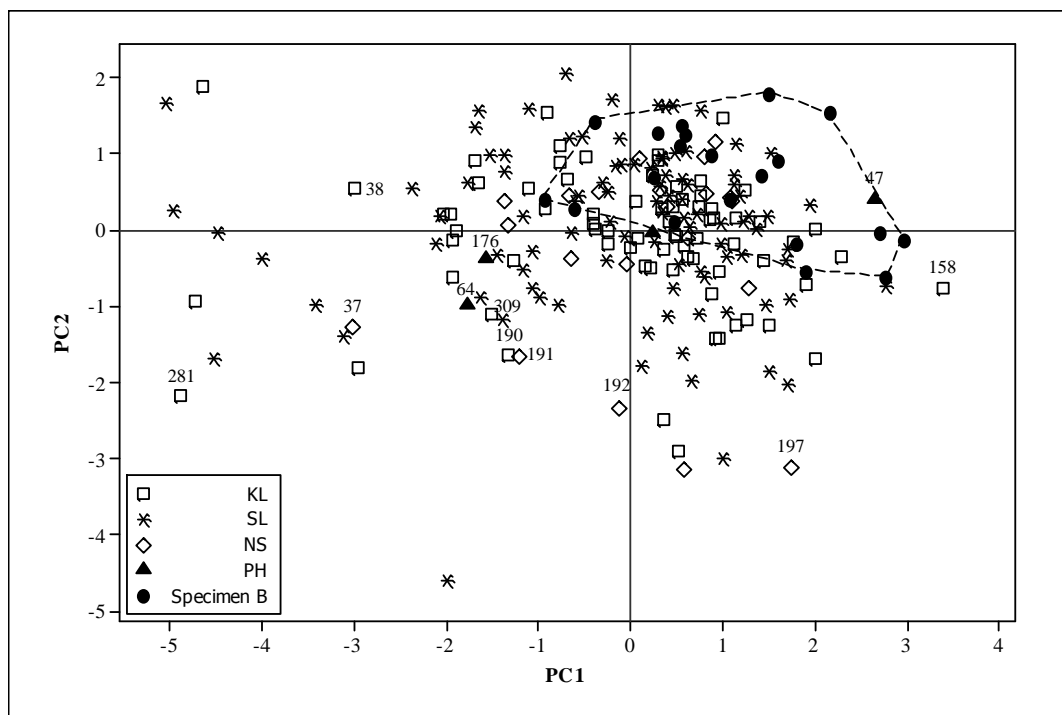


The data points of specimens B surrounded by a dotted polygon were included to serve as a control that collectively define the maximum natural deviation area (permitted instrumental and intra-batch variations) within which any deviations between the data points smaller than the polygonal area would suggest high similarity. Moving and rotating around the polygon on each score plot will help suggest if the spectral variation is due to natural variation or polymeric differences. The score plots in Figures 5.20(c) and 5.20(d) display poor similarities among the data points. This suggests that the plastic films used to pack the illicit heroin were likely to have originated from different batches/production lines at the distribution/street level.

Taking the spectral differences with or without width variation in the 197 cases into consideration, both score plots in Figure 5.21 show similar patterns. This indicates that an additional width variable in Figure 5.21(b) did not significantly change the general pattern on the first two components, although further separation (e.g. Film 192) or merging (e.g. Film 191) did occur in certain points. The spread of data was thus largely ascribed to the spectral differences. With the width variable, specimens B maintain their relatively random distribution with slight shifts in Figure 5.21(b). Based on the patterns, it could be concluded that the plastic packages used in the 197 cases were widely similar with some exceptions. Forensically, close agreements of the points (e.g. Films 190, 191, and 309) suggest that the same packer or different packers who purchased the same batch of plastic packets may be involved. In PH, Films 64 and 176 were unlikely to belong to the production line of Film 47 as they are marginally outside the maximum polygonal area. Film 38 was different from Film 158 although both were found in KL. Similarly, both Films 158 and 281 found in KL were noticeably different. It is also true in Films 37 and 197 seized in NS. Dissimilarities suggest different batches of plastic packets. This in turn implies that different packers or the same packer using different plastic films was involved.



(a)



(b)

Figure 5.21: Score plots representing IR data variation obtained from 197 Type 1 cases analyzed by PCA in correlation mode (The IR data were decomposed according to (a) normalized peaks only,  $\%V_1 = 57.28\%$  and  $\%V_2 = 26.73\%$ ; (b) normalized peaks and widths,  $\%V_1 = 46.20\%$  and  $\%V_2 = 21.44\%$ )

#### **5.3.4 Summary**

The polymer type of the plastic packets/bags remains a potential item for forensic intelligence. In this study, it was found that Type 1 plastic films were frequently used by heroin packers because of its easy accessibility and physical robustness for containing hard substances. The rare occurrence of Type 4 suggested a small scale operation. Associating the polymer types with information supplied by enforcement units can also provide a preliminary route for forensic intelligence. For spectral variation analysis, 197 small plastic packets (Type 1) were chosen. PCA was applied to the normalized spectral data in order to express the relationships between these plastic films at the distribution/street level. Based on the general data distribution pattern, most of the plastic packets were somewhat similar. This preliminary information is very useful for providing a gross overview of the relationships between the heroin cases at the street level.