Article

Prediction of Trained Panel Sensory Scores for Beef with Non-Invasive Raman Spectroscopy

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Abstract: The objective of this study was to investigate Raman spectroscopy as a tool for the prediction of sensory quality in beef. Raman spectra were collected from *M. longissimus thoracis et lumborum* (LTL) muscle on a thawed steak frozen 48 h post-mortem. Another steak was removed from the muscle and aged for 14 days before being assessed for 12 sensory traits by a trained panel. The most accurate coefficients of determination of cross validation (R^2 CV) calibrated within the current study were for the trained sensory panel textural scores; particularly *tenderness* (0.46), *chewiness* (0.43), *stringiness* (0.35) and *difficulty to swallow* (0.33), with practical predictions also achieved for *metallic flavour* (0.52), *fatty after-effect* (0.44) and *juiciness* (0.36). In general, the application of mathematical spectral pre-treatments to Raman spectra improved the predictive accuracy of chemometric models developed. This study provides calibrations for valuable quality traits derived from a trained sensory panel in a non-destructive manner, using Raman spectra collected at a time-point compatible with meat management systems.

Keywords: Raman spectroscopy; chemometrics; beef quality; trained sensory panel; tenderness

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1. Introduction

Sensory characteristics have a major influence on consumer eating satisfaction with regard to repurchasing fresh meat [1,2]. The most direct and accurate method of evaluating the sensory attributes of meat is through the use of trained panels [3,4]. However, sensory panels are expensive and time consuming to conduct [5]. Consequently, these methods are difficult to implement for either routine quality monitoring within meat management systems by commercial meat processors [6], or large scale, industry-wide recording of beef sensory data for the purposes of genetic evaluations [7].

Raman spectroscopy is a non-invasive vibrational spectroscopic technique that has applications in prediction of food quality. Raman spectroscopy measures the light scattered inelastically resulting from the interaction of a laser light with the molecules of a sample. Inelastic scattered light consists of photons that have experienced an energy/wavelength shift with respect to the incident light due to molecular vibrations. Therefore, the energy/wavelength shift is characteristic of the molecules present and can be used as a 'chemical fingerprint' of a sample [6]. Raman spectroscopy has numerous practical advantages in the context of food quality monitoring, including rapid measurement time, simple preparation of sample and non-destructive analysis, while providing high resolution detailed spectral information [3,8,9]. Another major advantage of Raman spectroscopy is its suitability for on-line implementation due to the use of fibre optics [10]. Moreover, it is relatively insensitive to water (in comparison to other technologies such as visible and near

infrared spectroscopy), rendering it suitable for application in the modelling/prediction of meat-eating quality traits [11].

The available literature is sparse in regard to the prediction of meat-trained sensory panel scores using Raman spectroscopy [4,8,9]. However, Raman spectroscopy has demonstrated its relevance to many areas of meat research. For example, it has been used successfully for the evaluation of meat spoilage [12,13], prediction of ultimate pH in pork [13], prediction of pork IMF content [14] and instrumental tenderness, IMF, cook-loss% and drip-loss% in beef [15,16]. Based on the demonstrated potential of Raman to capture the chemical signals associated with animal fat in terms of intramuscular fat and variability in ion content (in terms of pH in meat), and given the chemical nature of at least some sensory traits, we hypothesise that Raman spectroscopy would have potential for predicting meat eating quality. The objective of this study was to assess the potential of Raman spectroscopy as a tool for in-factory rapid prediction of meat eating quality. To this end, we collected non-destructive spectra on 2-day-aged beef samples and trained sensory panel scores from 14-day-aged beef samples from the same muscle, and conducted chemometric modelling of Raman spectra in relation to a range of sensory traits.

2. Materials and Methods

2.1. Animals and Sample Preparation

Crossbred bull and steer progeny ($n = 595$, 18 ± 4 months old) samples were collected [17]. Based on capturing maximal near infrared spectral variability within this data set [7], a subset of these animals ($n = 110$) were selected for sensory and Raman spectral analysis.

At 24 h post-mortem carcasses were quartered, and at 48 h post-mortem loins were boned out. Twelve steaks with a thickness of 2.54 cm were sliced from the right-side *LTL* starting at the anterior end, and vacuum packed. These steaks were labelled 1–12 in order of cutting. This step was included to ensure measurements were collected at a consistent location on the loin each time. The 5th and 6th steak removed from the loin were used for trained sensory panel analysis, while the 12th steak was allocated for Raman spectroscopy.

Samples allocated for Raman spectroscopy analysis were blast frozen at $-20\text{ }^{\circ}\text{C}$ and stored frozen until Raman spectra acquisition. Samples allocated for sensory analysis were aged in vacuum bags to 14 days post-mortem in a chill room set at $4\text{ }^{\circ}\text{C}$, then blast frozen at $-20\text{ }^{\circ}\text{C}$ and stored frozen until sensory analysis.

2.2. Raman Spectral Measurements

Prior to collection of spectra, *LTL* samples were thawed in a circulating water bath at room temperature. Once thawed, steaks were removed from the water bath, removed from vacuum packaging and lightly blotted with paper to remove excess surface moisture. Steaks were then wrapped tightly with oxygen-permeable clingfilm wrap, taking care not to trap air bubbles between the clingfilm wrap and *LTL* sample surface. Raman spectra were collected using a DXR SmartRaman spectrometer (ThermoFisher Scientific UK Ltd., Loughborough, UK) equipped with a diode laser operating at 780 nm, a charge-coupled device (CCD) detector and a universal platform sampling (UPS) accessory with a slit-aperture of $50\text{ }\mu\text{m}$. The muscle was scanned on the transverse surface of the *LTL*, in 10 representative locations (5 scans per side) within the muscle free from excessive fat and connective tissue. Raman intensity counts per second were recorded over the $250\text{--}3381\text{ cm}^{-1}$ Raman shift range with 2 cm^{-1} intervals via OMNIC for Dispersive Raman 9.2.98 software (ThermoFisher Scientific UK Ltd., Loughborough, UK). The scanning settings employed were: 120 mW laser power, 15s exposure time and 10 exposures. Cosmic spikes were automatically removed by the software.

2.3. Sensory Analysis

Sensory analysis was conducted at Teagasc Ashtown (Dublin 15, Ireland), following a modified version of the American Meat Science Association Research Guidelines for Cookery, Sensory Evaluation and Instrumental *Tenderness* Measurements of Meat [18].

Twelve sensory traits were scored (*tenderness, juiciness, chewiness, stringiness, crumbly texture, fatty mouthfeel, difficulty to swallow, beef flavour, metallic flavour, beef after-effect, metallic after-effect and fatty after-effect*) on a 0–100 line scale, with 0 being the lowest score and 100 the highest. A detailed description of this method is provided in Cafferky et al. [7].

2.4. Chemometric Analysis

Spectral pre-processing and data analysis were performed with the statistical application R (R Core Team, 2018) using the R packages, (baseline, signal, EMSC, pls, plsVarSel and other in-house functions). Calculations were based on Raman shift ranges from 3100 to 700 cm^{-1} , as a low signal-to-noise ratio was observed outside this range. Two ranges were evaluated: 3100–700 cm^{-1} and 1800–700 cm^{-1} . For the prediction of beef sensory attributes, partial least squares regression (PLSR) models were developed for each of the 12 sensory and textural traits analysed (Y data), using Raman spectra collected from intact *LTL* samples (X data), with 104 models constructed in this study in total. Variable importance on projection (VIP) was employed as the algorithm of choice for variable selection [19]. VIP was conducted with a 1 threshold and iteratively (backwards variable elimination) until the minimum root mean square error of cross validation (RMSECV) was obtained. Four spectral pre-treatments were evaluated in the current study as follows: Extended multiplicative scatter correction (EMSC); Savitzky-Golay first derivative, second-degree polynomial with nine smoothing points (SG1); Savitzky-Golay second derivative, second-degree polynomial with nine smoothing points (SG2); and baseline correction (polynomial of 5th order). PLSR models were calibrated using the kernel algorithm, with the performance of models evaluated using twenty iterations of a ten-fold cross validation. Parameters examined in model performance evaluation included coefficient of determination of calibration (R^2C), coefficient of determination of cross validation (R^2CV), root mean square error of calibration (RMSEC) and root mean square error of cross-validation (RMSECV).

3. Results and Discussion

3.1. Descriptive Statistics of Sensory Traits

Descriptive statistics for the 12 beef sensory traits are presented in Table 1. Large coefficients of variation were present in the textural sensory traits of chewiness, crumbliness, stringiness and difficulty to swallow (CV 46.5% to 80.8%); and also for the flavour-related traits of metallic flavour, metallic after-effect, fatty mouthfeel and fatty after-effect (CV 40.8% to 77%). Lower variability among samples was observed for juiciness, tenderness, beef flavour and beef after-effect (17.5% to 23.1%).

Table 1. Descriptive statistics of twelve beef *LTL* eating quality traits (sensory scores measured on a scale of 0–100).

	Mean	SD	Median	Min	Max	CV
<i>Tenderness</i>	51.85	11.98	54.91	23.25	70.75	23.1%
<i>Chewiness</i>	26.98	12.55	23.35	6.83	56.33	46.5%
<i>Stringiness</i>	12.31	9.95	8.81	0.43	40.94	80.8%
<i>Difficulty to swallow</i>	12.25	8.09	10.14	0.83	33.13	66.0%
<i>Crumbliness</i>	20	11.21	17.63	2.33	45.3	56.1%
<i>Beef flavour</i>	41.16	7.22	41.27	20.6	60.88	17.5%
<i>Beef AE</i>	31.2	5.76	31.23	16.5	45	18.5%
<i>Juiciness</i>	33.51	7.06	33.2	17.5	54.35	21.1%
<i>Fatty mouthfeel</i>	5.96	4.56	4.63	0.21	19.63	76.5%
<i>Fatty AE</i>	6.38	4.91	5	0.5	24.25	77.0%
<i>Metallic flavour</i>	13.91	5.9	12.77	1.42	29.5	42.4%
<i>Metallic AE</i>	18.49	7.54	18.65	3.67	37.63	40.8%

SD, Standard Deviation; Min, Minimum; Max, Maximum; CV, Coefficient of Variation; AE, After-effect; Sensory scores are measured on a scale of 0–100.

3.2. Prediction of Beef *LTL* Sensory Tenderness by Raman Spectroscopy

To evaluate the potential of Raman spectroscopy for prediction of beef sensory quality, PLSR models were developed for each sensory trait using various pre-processing techniques

and Raman shift ranges. Models based on the Raman shift range from 1800 to 700 cm^{-1} resulted in increased predictive ability. This effect was attributed to the fact that the range from 1800 to 500 cm^{-1} contains most of the relevant Raman scattering responses of meat [16]. Average spectra for the five highest and five lowest performing samples for tenderness, chewiness and crumbliness may be seen in Supplementary file Figure S1.

The best-fitting prediction models for each beef sensory and textural traits using Raman spectroscopy are presented in Table 2. It is widely considered that *tenderness* is the most important sensory trait for consumers, with first bite tenderness the basis on which many consumers make a decision on whether they will re-purchase [20], while at the same time consistently being the trait most difficult to predict without destruction of the sample [9]. Best-fitting models for prediction of *tenderness* gave moderate results ($R^2\text{CV} = 0.46$; $\text{RMSECV} = 8.74$). EMSC was found to be more effective than SG pre-processing for prediction of *tenderness*, with both models calibrated using two latent variables (LV). Indeed, the best fitting models for all traits presented in the current study were all calibrated within the first two LV.

Table 2. Best fitting predictions of twelve sensory traits on beef *LTL* using Raman spectral measurements.

Variable	Math Treatment	Var	P	$R^2\text{Cal}$	RMSEC	$R^2\text{CV}$	RMSECV
Tenderness	EMSC	131	2	0.66	6.92	0.46	8.74
	SG2	265	2	0.61	7.41	0.33	9.74
Chewiness	EMSC	119	2	0.64	7.54	0.43	9.46
	SG2	255	2	0.59	8.05	0.34	10.15
Stringiness	EMSC	120	1	0.46	7.31	0.35	7.96
	SG1	69	3	0.46	7.27	0.35	7.97
Diff. swallow	SG2	270	2	0.61	5.03	0.33	6.59
	EMSC	358	1	0.51	5.67	0.33	6.62
Crumbliness	EMSC	126	1	0.45	8.27	0.36	8.92
	SG2	244	2	0.49	7.96	0.31	9.28
Beef flavour	EMSC	149	1	0.36	5.77	0.22	6.36
	SG2	286	2	0.54	4.87	0.16	6.60
Beef AE	EMSC	391	1	0.42	4.35	0.27	4.91
	SG2	233	2	0.53	3.92	0.14	5.32
Juiciness	SG2	275	2	0.60	4.43	0.36	5.63
	EMSC	132	2	0.60	4.46	0.30	5.88
Fatty mouthfeel	EMSC	353	1	0.51	3.19	0.34	3.70
	SG2	286	2	0.58	2.94	0.31	3.76
Fatty AE	EMSC	338	1	0.41	2.90	0.23	3.31
	SG2	265	2	0.70	2.08	0.44	2.82
Metallic flavour	SG2	258	2	0.70	3.25	0.52	4.08
	EMSC	128	1	0.45	4.35	0.35	4.74
Metallic AE	EMSC	128	1	0.5	5.33	0.37	5.94
	SG2	212	2	0.54	5.08	0.28	6.37

Var, number of spectral variables retained by Variable Importance in Projection; P , number of PLS terms utilised in the calibration of the equation; $R^2\text{C}$, coefficient of determination of calibration; RMSEC, root mean square error of calibration; $R^2\text{CV}$, coefficient of determination of cross validation; RMSECV, root mean square error of cross validation; Diff. Swallow, Difficulty to swallow; AE, after-effect; EMSC, Extended multiplicative scatter correction; SG1, Savitzky-Golay first derivative, second-degree polynomial with nine smoothing points; SG2, Savitzky-Golay second derivative, second-degree polynomial with nine smoothing points.

The correlation between Raman spectra and *tenderness* scores (trained sensory panel) was evaluated based on the loading plot values obtained for PLS factors 1 and 2 (Figure 1a). This plot provides information on the Raman bands that contributed to each PLS factor [21]. The plot identified a series of bands that contribute to the prediction of *tenderness*, most of which are related to β -sheet folding; an example of this are the bands from 1680 to 1665 cm^{-1} [3]. Both PLS factors 1 and 2 indicate that this region is positively correlated to tenderness. This is noteworthy, since higher prevalence of β -sheet structures in meat are associated with toughness, rather than tenderness [3,22]. Yet, higher intensity at the region 1685–1645 cm^{-1} may be due to contribution from lipids such as the C=C stretching mode occurring around 1680–1640 cm^{-1} . Higher fat content has been previously associated with an increase in meat tenderness. PLS factor 2 also indicates that the band situated at 962 cm^{-1} , which is associated with C–C stretching vibrations of random coil, is negatively correlated

to *tenderness* scores [3,22]. Furthermore, PLS factor 1 indicates that the band at 1263 cm^{-1} , which may be attributed to the presence of connective tissue [15], has a negative correlation to *tenderness*. Other prominent bands correlated to *tenderness* scores include the 1322 and 1354 cm^{-1} band related to asymmetrical SO_2 stretching and the bands visible at 1605 , 1617 , 1658 and 1784 cm^{-1} , which are located within the primary thioamides-Amide II band [23].

The samples analysed in the current study (for the prediction of trained sensory panel scores for beef via Raman spectroscopy) are a subset of a larger group of samples analysed in a study investigating the use of visible–near infrared spectroscopy (VISNIRS) for the prediction of trained sensory panel scores for beef [7]. When comparing our previous predictions of sensory *tenderness* using VISNIRS to Raman spectroscopy, Raman spectroscopy predictions outperform VISNIRS prediction in nearly every parameter (VISNIRS $R^2\text{CV} = 0.13$; RMSECV = 8.6; LV = 6).

This study is novel in the context of Raman prediction of *tenderness*, because we recorded spectra on uncooked, intact muscle, at an early time point post-mortem. This makes the process highly relevant to the need for rapid online predictions characteristic of the meat industry, which rarely have the facilities to age, cook and/or homogenise samples for the prediction of sensory attributes prior to sale. Beattie et al. [3] developed models for trained sensory panel *tenderness* scores of beef silverside, however in that study, spectral collection and sensory panel analysis were both carried out on 21-day-aged and cooked beef. While the prediction was slightly more accurate in that study ($R^2 = 0.65$), it came with a larger error value (RMSECV = 9.69). Furthermore, our rapid prediction at 2 days post-mortem would be more relevant for industrial application in terms of furthering rapid technologies to aid meat management strategies and animal breeding for meat quality. The predictive ability of the model for beef *tenderness* presented in the current study ($R^2\text{CV} = 0.46$), which was found on meat aged for only 2 d, has superior potential for integration into routine process analytical monitoring in meat plants. Zhao et al. [9] reported a similar prediction for *tenderness* ($R^2\text{CV} = 0.45$) to our study, but they calibrated models using Raman spectra collected from homogenised beef samples. Our study demonstrates that the predictive accuracy of models calibrated using Raman spectra for *tenderness* is not improved through meat homogenisation (though this may be more relevant for traits associated with fat content, such as *juiciness* and *flavour*).

(a)

Figure 1. Cont.

(b)

(c)

Figure 1. Cont.

(d)

(e)

Figure 1. Cont.

(f)

(g)

Figure 1. Cont.

(h)

(i)

Figure 1. Cont.

(j)

(k)

Figure 1. Cont.

(I)

Figure 1. Loading values of first/first two PLS factors employed by the selected models for (a) *tenderness*, (b) *chewiness*, (c) *stringiness*, (d) *difficulty to swallow*, (e) *crumbliness*, (f) *juiciness*, (g) *beef flavour*, (h) *beef after-effect*, (i) *fatty mouthfeel*, (j) *fatty after-effect*, (k) *metallic flavour*, (I) *metallic after-effect*.

3.3. Prediction of Other Beef LTL Textural Traits by Raman Spectroscopy

Traditionally, trained beef sensory panels only score samples on traits such as *tenderness*, *flavour* and *juiciness*, with the occasional exception of *chewiness* included. With the insight and feedback of the trained sensory panel (after undergoing initial training and screening), the number of traits to be analysed was expanded (particularly in regards to texture) in an effort to subdivide the entire sensory profile of our samples to try to provide novel and more refined predictions that the wider meat industry could use in the future for more detailed selection for meat quality.

The textural sensory traits most commonly associated with toughness—*chewiness*, *stringiness* and *difficulty to swallow*, were also predicted with reasonable accuracy (Table 2). Of these three traits, *chewiness* was predicted with the highest accuracy ($R^2CV = 0.43$; $RMSECV = 9.46$; $LV = 2$). *Difficulty to swallow* and *stringiness* models calibrated using Raman spectra gave similar results ($R^2CV = 0.33$; 0.35), with the *difficulty to swallow* model having slightly lower $RMSECV$ values (6.62 and 7.96). Both models were calibrated in the first PLS term ($LV = 1$), suggesting that they were robust and repeatable.

In comparison to previous predictions of sensory meat quality traits using VISNIRS by our group [7], Raman spectroscopy outperformed VISNIRS in traits related to texture (VISNIRS R^2CV *difficulty to swallow* = 0.1; *chewiness* = 0.17; *stringiness* = 0.22). Models calibrated using VISNIRS were also calibrated using more PLS terms (LV 9 and 10), indicating that the Raman models calibrated in the current study may be more repeatable and robust. Zhao et al. [9] reported predictions calibrated using Raman for trained sensory panel *chewiness*, *stringiness* and *cohesiveness* (comparable to *difficulty to swallow*) scores with greater accuracy ($R^2CV = 0.63$; 0.63 ; 0.64) and lower error value ($RMSECV = 8.48$; 2.98 ; 5.93) than the current study, suggesting that homogenisation of the tissue (and subsequent destruction of muscle fibre formation) may improve the predictive ability of traits related to sensory toughness. Examination of the loading plots presented in Figure 1b, 1c and 1d identified a number of bands that contribute most to the prediction of tough sensory traits. *Chewiness* (1661 , 1681 cm^{-1}), *stringiness* (1680 cm^{-1}) and *difficulty to swallow* (1666 cm^{-1}) were all prominent bands that positively correlate with β -sheet folding (with tougher meat having a greater amount of β -sheet structures) as discussed by Beattie et al. [3].

Raman predictions for *crumbliness* gave similar model performance to the other textural traits previously discussed ($R^2CV = 0.36$; $RMSECV = 8.92$). This model was constructed with the use of one LV and EMSC spectral pre-processing. While the predictive accuracy of the models is not very high, the fact that the model is in the first term indicates that it is quite stable. In previous research carried out by this group, *crumbliness* was predicted with VISNIRS with higher predictive accuracy ($R^2CV = 0.41$) and comparable $RMSECV$ value (9.4); however, seven LV were needed to calibrate the best fitting model [7]. When comparing the two technologies, taking the slight difference in R^2CV value and the large difference in number of LV used to calibrate the models, the data suggests that Raman spectroscopy may be a more repeatable and robust method of spectroscopy for the prediction of *crumbliness*.

3.4. Prediction of Beef LTL Sensory Flavour and Juiciness by Raman Spectroscopy

Best-fitting predictions for *beef flavour* provided the least accurate models, having an R^2CV value of 0.22 and an $RMSECV$ value of 6.36 (1 LV) (Table 2). Previously, models were calibrated for *beef flavour* prediction by our group using VISNIRS. Unsatisfactory predictions with VISNIRS were found for this trait ($R^2CV = 0.13$; $RMSECV = 6.4$) [7], suggesting that spectroscopy on the intact muscle is not a useful mechanism to predict flavour.

Beef after-effect predictive models were slightly more accurate than *beef flavour* models in terms of higher R^2CV (0.27) and lower $RMSECV$ value (4.91), with the same number of LV used in calibration (one), indicating superior model robustness and repeatability.

The best fitting model for sensory panel *juiciness* scores predicted via Raman spectra gave an R^2CV of 0.36 and a relatively low $RMSECV$ of 5.63, constructed using two LV (Table 2). Interestingly, *juiciness* is one of only three traits in the current study where SG smoothing provided a more accurate model than EMSC. Fowler et al. [6] presented a similar predictive value for *juiciness* using an untrained sensory consumer panel ($R^2CV = 0.42$), however with a much higher margin of error ($RMSECV = 11.29$) and with the use of more PLS terms (LV = 5).

In the current study, models for the prediction of *fatty mouthfeel* and *fatty after-effect* were calibrated with a moderate degree of success (Table 2). Best fitting models for *fatty mouthfeel* provided an R^2CV of 0.34 ($RMSECV = 3.7$; 1 LV). When comparing VISNIRS to Raman spectroscopy for the prediction of this trait, the best-fitting VISNIRS model only predicted *fatty mouthfeel* with an R^2CV of 0.23 [7].

Fatty after-effect was predicted slightly more accurately in the current study than *fatty mouthfeel* ($R^2CV = 0.44$; $RMSECV = 2.82$; 2 LV). *Fatty after-effect* is predicted with increased efficacy using Raman spectroscopy relative to VISNIRS as shown in [7], where the best-fitting model had a lower R^2CV (0.28), increased $RMSECV$ (3.2) and took 7 LV to calibrate the predictive model, indicating that the Raman model is the more robust and superior of the two.

The relationship between Raman spectra and trained sensory panel *fatty mouthfeel* and *fatty after-effect* scores are presented in Figure 1i and 1j by evaluating the loading plot values obtained for PLS factors 1 and 2. With respect to *fatty mouthfeel*, pronounced bands occur at 1680 cm^{-1} (Amide I), 1271 cm^{-1} (Amide III) and 1062 cm^{-1} , 1097 cm^{-1} , 1122 cm^{-1} and 1130 cm^{-1} (C-C stretching modes) [23], while for *fatty after-effect*, bands are prominent at 1667 cm^{-1} and 1659 cm^{-1} (Amide I), 1271 cm^{-1} (Amide III) and 1032 cm^{-1} , 1050 cm^{-1} , 1073 cm^{-1} and 1103 cm^{-1} [23]. Contribution of the region $1685\text{--}1645\text{ cm}^{-1}$ may be also associated with lipids such as the C=C stretching mode occurring around $1680\text{--}1640\text{ cm}^{-1}$ [22]. Similarly, the intensity increase at 1130 cm^{-1} could be associated with C-C stretching modes of lipids [6].

Metallic flavour and *metallic after-effect* were positively correlated to each other [7], however in this study, the two traits were predicted with differing accuracies. *Metallic flavour* was more accurately predicted ($R^2CV = 0.52$; $RMSECV = 4.08$; two LV), compared to *metallic after-effect* ($R^2CV = 0.37$; $RMSECV = 5.94$; one LV). This is not surprising, as sensory after-effects are by nature indirect and slightly removed from the actual eating experience. The prediction

value achieved for this trait was three-fold greater than the result achieved using VISNIRS ($R^2CV = 0.17$; $RMSECV = 4.38$; 10 LV), with eight fewer LV needed to calibrate a model. Indeed, even for *metallic after-effect*, the predictive models we obtained via Raman spectra were better than those obtained previously using VISNIRS ($R^2CV = 0.19$; $RMSECV = 6.6$; 10 LV) [7]. In Figure 1k, positive correlations can be seen between *metallic flavour* trained sensory panel scores and bands situated at 829, 853, 1002, 1128, 1450 and 1464 cm^{-1} , while negative correlations exist between *metallic flavour* and the bands 842, 998, 1009, 1140, 1215 and 1432 cm^{-1} . Correlations between Raman spectral bands and *metallic after-effect* are presented in Figure 1l, with strong positive correlations between the trait and the bands at 885, 1107, 1199, 1294 and 1480 cm^{-1} . *Metallic after-effect* was negatively correlated with the bands 1066, 1117, 1982, 1371, 1733 and 1749 cm^{-1} . When comparing our results for prediction of beef sensory *flavour* and *juiciness* via Raman spectroscopy to the most similar (yet less industrially relevant in design) published studies, the predictions (for the most part) are within a similar range. Zhao et al. [9] presented predictions that were more accurate than the current study for the sensory traits; beef flavour, beef flavour length (comparable to *beef after-effect*), residual metallic (comparable to *metallic after-effect* in the current study) and *fatty mouthfeel* (comparable to the fattiness trait presented) ($R^2CV = 0.61$ – 0.84 ; $RMSECV = 2.31$ – 4.65); whereas Beattie et al. [3] predicted beef flavour in a similar fashion to the current study ($R^2CV = 0.26$; $RMSECV = 7.74$). Predictions obtained in this study for *fatty after-effect* were in the same range as the result obtained by Zhao et al. [9] for the comparable residual fattiness sensory trait ($R^2CV = 0.54$; $RMSECV = 2.42$). However, the predictions of *metallic flavour* in both studies are very similar ($R^2CV = 0.54$; $RMSECV = 2.55$). While the homogenisation of samples may aid the prediction of beef sensory flavour traits by Raman spectroscopy (e.g., homogenising fat, protein and flavour compounds within the sample), when looking across the full range of sensory flavour traits, decent predictions can be obtained without the need for homogenisation, leading to a more rapid, non-destructive technology with greater scope for the meat industry.

4. Conclusions

The results of this research demonstrate that Raman spectra collected from beef *LTL* samples have the potential to predict a wide range of trained sensory panel scores with varying levels of accuracy and margin of error, dependent on the trait. The most accurate coefficients of determination of cross validation calibrated within the current study were for the trained sensory panel textural scores; in particular *tenderness*, *chewiness* and *stringiness*, with practical predictions achieved for *metallic flavour*, *fatty after-effect* and *juiciness*. EMSC spectral pre-processing provided the best-fitting model for all but one trait evaluated in this study. Raman spectroscopy is a valuable tool for breeders and processors with regard to the prediction of beef quality as the spectra can be recorded non-destructively and rapidly, with potential for on-line application. Information can be captured on wide ranging and difficult to measure traits, once the calibrations are available. This study provides calibrations for valuable quality traits derived from a trained sensory panel in a non-destructive manner, using Raman spectra collected at a time-point compatible with meat management systems, and hence may have relevance to future breeding programmes for beef sensory quality.

Supplementary Materials: The following supporting information can be downloaded at: <https://www.mdpi.com/article/10.3390/chemosensors10010006/s1>, Figure S1: Average spectra for five highest and lowest scoring samples for each of three sensory traits (a) tenderness, (b) chewiness and (c) crumbli-ness. Less favourable profiles in red and more favourable profiles in dashed blue for tenderness and crumbli-ness, while for chewiness, less favourable profiles are in dashed blue and more favourable profiles are in red.

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