



final report

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Beef shelf life prediction model

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Modelling shelf life of MAP Packed Beef (80 % Carbon dioxide + 20 % Oxygen)

Executive Summary

The beef industry has a challenge in improving and documenting shelf life of chill stored beef along the distribution chain. This project aims to adapt the existing Danish Meat Research Institute (DMRI) shelf life prediction model for vacuum packed product to retail packing in modified atmosphere packaging (MAP). A model describing growth and development of raw meat odour in MAP packed beef has been developed.

The difference between the counts found and scores for raw meat odour during storage of MAP packed (80 % CO₂ and 20 % O₂) beef is not very different from what would be expected in vacuum packed beef. There is a tendency, that the difference is larger at low temperatures than at high temperatures in the range -0.3 °C to 8 °C. The following cuts were used for the tests:

- Topside with fat
- Cuvette - a Scandinavian cut from the hind leg suitable for production of minced meat
- Sirloin

The count N at the time t is calculated with the formula below, where N_0 is the count at time 0, N_{∞} is the maximum count, and μ_{\max} is the max growth rate.

$$\ln(N) = \ln(N_0) + \mu_{\max} A(t) - \ln \left(1 + \frac{e^{\mu_{\max} A(t)} - 1}{(N_{\infty} - N_0)} \right)$$

$$A(t) = t + \frac{1}{\mu_{\max}} \ln \left(e^{-h} + (1 - e^{-h}) e^{-\mu_{\max} t} \right)$$

Where h is a constant determining the lag phase.

The relationship between μ_{\max} and temperature can be described with the formula below, where β_0 and β_1 are constants.

$$\mu_{\max} = \beta_0 e^{\beta_1 T}$$

The constants determined for this model were:

	Estimate	Approx. Std. Error	Approx. 95% Confidence limits	
β_0	0.00606	0.00163	0.00276	0.00935
β_1	0.1933	0.0303	0.1322	0.2544
h	-1.6602	1.1424	-3.9657	0.6454

RMSE (Root Mean Square Error) when comparing fitted and found values is approx. 0.8 and 1.2 for growth and development in raw meat odour respectively. When developing the model for shelf life of vacuum packed beef an RMSE of 0.38 and 0.78 were found for growth and raw meat odour. This

means that the differences between observed and fitted values are larger in the model for MAP packed beef than in the model for vacuum packed beef.

This model will be incorporated into a Shelf Life Prediction Tool for the Australian red meat industry.

Summary

Background

The beef industry has a challenge in improving and documenting shelf life of chill stored beef along the distribution chain. DMRI has developed a first step model to be used by the industry for prediction of shelf life. The model is developed with the aim to be as simple as possible, with few parameters, and with parameters that are either known or can be gathered at low cost.

Purpose

MLA has requested DMRI to conduct an adaption of the existing DMRI shelf life prediction model to Australian conditions, including a research study of extension modules in areas of repacking and retail packing in modified atmosphere packaging (MAP), 80 % CO₂ and 20 % O₂.

Conclusions

A model describing growth and development of raw meat odour in MAP packed beef has been developed. RMSE (Root Mean Square Error) when comparing fitted and found values is approx. 0.8 and 1.2 for growth and development in raw meat odour respectively. When developing the model for shelf life of vacuum packed beef an RMSE of 0.38 and 0.78 were found for growth and raw meat odour. This means that the differences between observed and fitted values are larger in the model for MAP packed beef than in the model for vacuum packed beef.

The difference between the counts found and scores for raw meat odour during storage of MAP packed beef is not very different from what would be expected in vacuum packed beef. There is a tendency, that the difference is larger at low temperatures than at high temperatures in the range -0.3 °C to 8 °C.

Introduction

Background

The beef industry has a challenge in improving and documenting shelf life of chill stored beef along the distribution chain. It is important for both consumer satisfaction and authority requirements to determine and document realistic use-by dates. Furthermore, it is important to communicate how the time temperature course affects shelf life in order to optimize the different steps on the way.

DMRI has developed a first step model to be used by the industry for prediction of shelf life, described in DMRI report doc 49409 and at ICOMST 2009 in manuscript: Meinert, L., Koch, A.G., Blom-Hanssen, J., Bejerholm, C., Madsen, N.T. & Christensen, H., 2009. Predicting the shelf-life of fresh pork and beef. Manuscript PE8.19.00, 55th International Congress on Meat Science & Technology, Copenhagen 16 - 21 August.

The model is developed with the aim to be as simple as possible, with few parameters, and with parameters that are either known or can be gathered at low cost. DMRI researched whether initial load and growth of preferably one microbial parameter could be used to predict shelf life. On the basis of this work, a deterministic model that describes the influence of bacteriology, storage temperature(s) and packing method(s) on the "organoleptic shelf life" of fresh beef has been developed. The model exists in a spreadsheet format for vacuum packed beef; the model has been successfully tested at several Scandinavian beef companies for documentation and improvement purposes.

Purpose

MLA has requested DMRI to conduct an adaption of the existing DMRI shelf life prediction model to Australian conditions, including a research study of extension modules in areas of repacking and retail packing in modified atmosphere packaging (MAP).

Experimental work - short description

Cuts (Topside, Cuvette, and Sirloin) were collected at commercial packing plants in Denmark and Germany and transported to DMRI's pilot plant facilities, where they were MAP packed and subsequently stored at 5 different temperatures. For each storage temperature, 5 packages were evaluated 8 - 11 times during the storage period. The number of psychrotrophs was determined on the cut in each pack. Upon opening of the packs, each cut was evaluated organoleptically (raw meat odour and visual appearance) and scored by at least 3 persons from a pool of "lay men". For raw meat odour the following scale was used - 2: fresh; 4: slightly diverging but acceptable; 6: diverging to an unacceptable degree; 8: putrid/rotten.

Materials and methods

Cuts

The following cuts were used for the tests:

- Topside with fat - purchased from a German packaging plant. Topsides were selected at random from the deboning belt in the plant (4 February 2010). Animals were of German origin and slaughtered one day before cutting and deboning.
- Cuvette - a Scandinavian cut from the hind leg. In most countries outside Scandinavia it is used for production of minced meat. Cuvettes were purchased at a Danish packaging plant. The cuts were selected at random during deboning of hind quarters (1 March 2010). The animals were of Danish origin and slaughtered 2 days before cutting and deboning.
- Sirloins - purchased from a German packaging plant. Sirloins were selected at random during deboning (23 March 2010). The animals were of German origin and slaughtered 2 days before deboning.

Cuts were transported under chilled conditions to DMRI's pilot facilities in Roskilde, Denmark.

Packaging

Upon arrival at DMRI (1 day after deboning) the cuts were divided into pieces of approx. 350 g and packed in sealed trays with a gas consisting of 80 % CO₂ and 20 % O₂, see Figure 1.

Trays were supplied by Færch Plast (<http://www.faerchplast.com/>) - Article no.: MAPET® K 71-51 W.

Film was also supplied by Færch Plast - Article no: Topseal® pet map pb 62. Datasheet is attached as Annex 12.

The gas was supplied by Yara Praxiair, and the concentration was certified to be within +/- 0.5 %.

Each time cuts were packed, the concentration of CO₂ and O₂ was measured in a number of empty packs to verify the gas composition at the time of packaging.

Gas composition

Gas composition was measured with a "Checkmate 9900" manufactured by PBI Dansensor.

pH

pH was measured in all samples for microbiological and sensory evaluation.



Figure 1: Packaging

*Microbiology - sampling
and analysis*

The sampling method was destructive. Samples were taken arbitrarily irrespective of surface type (fat, lean meat, membrane or combinations).

10 cm² of the surface was marked, removed and transferred to a stomacher bag. The bag was added 100 ml of Buffered Peptone Water and stomached for 1 min.

To obtain a microbial profile at the beginning of storage, 15 samples were taken upon arrival at DMRI and analyzed quantitatively for:

- Psychrotrophs - PCA - incubation at 6.5 °C for 10 days
- Psychrotrophs - IDF 132 (ISO 8552)
- Aerobic count - PCA - incubation at 20 °C for 3 days
- Lactic acid bacteria - MRS-agar - incubation at 20 °C for 3 days (NMKL no. 140, 2nd ed., 2007)
- Enterobacteriaceae - Violet Red Bile glucose agar - incubation at 37 °C for 1 day (NMKL no 144. 3rd ed. 2005) - NB: All colonies that grow on the media were counted.

For modelling purposes, 5 samples were drawn at random 8 - 11 times per storage temperature and analysed quantitatively for:

- Psychrotrophs - PCA - incubation at 6.5 °C for 10 days
- Psychrotrophs - IDF 132 (ISO8552)

Sensory evaluation

Upon opening of packs for microbial analysis, the packs were evaluated organoleptically - raw meat odour and visual appearance. Before the evaluation, the packages were left open to “aerate” at approx. 20 °C for at least 20 minutes.

The scoring was performed by at least 3 persons from a pool of employees at DMRI.

The following scales were used:

Raw Meat odour:

- 2 - Fresh
- 4 - Slightly diverging, but acceptable
- 6 - Diverging to an unacceptable degree
- 8 - Putrid / Rotten

Visual appearance:

- 1 - No discolouration
- 2 - Slightly discoloured, but acceptable
- 3 - Discoloured to an unacceptable degree
- 4 - Very discoloured

Statistics and modelling

All microbial counts were reported as log cfu/cm².

Fitting of both the microbial growth curves and the raw meat odour was performed using nonlinear regression (proc nlin, SAS®, version 9.2 TS level 2M0) with the growth model developed by Baranyi and Roberts (J. Baranyi and T. A. Roberts. "A dynamic approach to predicting growth in food". Int. J. of Food Microbiology. Vol. 23 1994, pp. 277 - 294).

The count N at the time t is calculated with the formula below, where N_0 is the count at time 0, N_∞ is the maximum count, and μ_{\max} is the max growth rate.

$$\ln(N) = \ln(N_0) + \mu_{\max} A(t) - \ln \left(1 + \frac{e^{\mu_{\max} A(t)} - 1}{e^{\mu_{\max} A(0)} - 1} \right)$$

$$A(t) = t + \frac{1}{\mu_{\max}} \ln \left(e^{-h} + (1 - e^{-h}) e^{-\mu_{\max} t} \right)$$

Where h is a constant determining the lag phase.

The relationship between μ_{\max} and temperature can be described with the formula below, where β_0 and β_1 are constants.

$$\mu_{\max} = \beta_0 e^{\beta_1 T}$$

The same formulas as above are used to fit the "raw meat odour" curves with the exception that h is calculated on the basis of N_0 and a constant called $\log(N_{lugt})$.

$$h = \log(N_{lugt}) - \log(N_0)$$

For each fit RMSE (Root Mean Square Error) has been calculated with the formula below.

$$RMSE = \sqrt{\frac{\sum_{i=1}^n (\text{Predicted} - \text{observed})^2}{n}}$$

Where n is the number of observations.

Results and discussion

Temperatures during storage

Temperatures were logged during storage. The results were as follows. The first figure is the average temperature in the room, and the numbers in brackets are the average and standard deviation for each logger.

- -0.3 °C (-0.43 +/- 0.27; -0.27 +/- 0.33; -0.23 +/- 0.17) - Topsides
- 1.2 °C (1.25 +/- 0.39; 1.10 +/- 0.39) - Cuvetter
- 3.4 °C (3.08 +/- 0.32; 3.63 +/- 0.34) - Cuvetter
- 5.9 °C (6.1 +/- 0.99; 6.04 +/- 1.17) - Sirloins
- 8.1 °C (8.12 +/- 0.08; 8.09 +/- 0.09; 8.09 +/- 0.08) - Topsides

The average temperatures are used in the following.

Microbial profiles at the beginning of storage

Average counts at the beginning of storage are shown graphically in Figure 2 and in a tabular form in Annex 1.

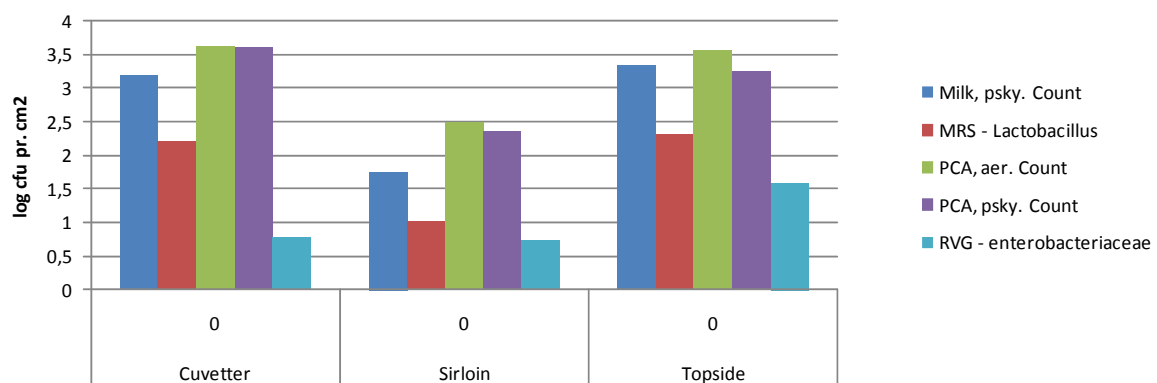


Figure 2: Average counts at the beginning of storage, log cfu/cm². n=15 per bar. "Milk, psky count": Psychrotrophs - IDF 132 (ISO8552). "MRS - Lactobacillus": Lactic acid bacteria on MRS-agar. "PCA, aer. Count": Aerobic count - incubation on PCA at 20 °C for 3 days. "PCA, psky count": Psychrotrophs - PCA - incubation at 6.5 °C for 10 days. "RVG - enterobacteriaceae": Enterobacteriaceae - incubation on RVG at 37 °C for 1 day.

The number of *enterobacteriaceae* on Cuvetter, Sirloins and Topsides were below the detection limit (10 cfu per cm²) in 12, 13 and 5 samples (n=15). When calculating the average, the number was set to 0.7 log cfu/cm² (5 cfu per cm²).

The initial counts on Cuvetter and Topsides were fairly similar with the exception of the number of *enterobacteriaceae* which was higher on Topsides, than on Cuvetter. The level of all parameters was lowest on Sirloins.

Changes in raw meat odour and visual appearance during storage

At all 5 temperatures the tests “were running” until all of 5 samples drawn on a specific day were sensory unacceptable twice in a row; i.e. the score for raw meat odour was ≥ 6 for all samples and/or the score for visual appearance was ≥ 3 for all samples.

Results from storage at -0.3°C and at 8.1°C are shown in figure 3 and figure 4. Results from all storage temperatures are shown in a tabular form in Annex 2.

After the end of shelf life, the odour of individual MAP packed cuts of beef differs from each other and very rarely turns rotten/putrid. The odour developing after the end of shelf life can be characterized with words/phrases like: “very unpleasant in a way hard to define”, “old”, “sour”. This is contrary to the development of the odour in vacuum packed beef, where the odour almost always turns rotten/putrid after the end of shelf life.

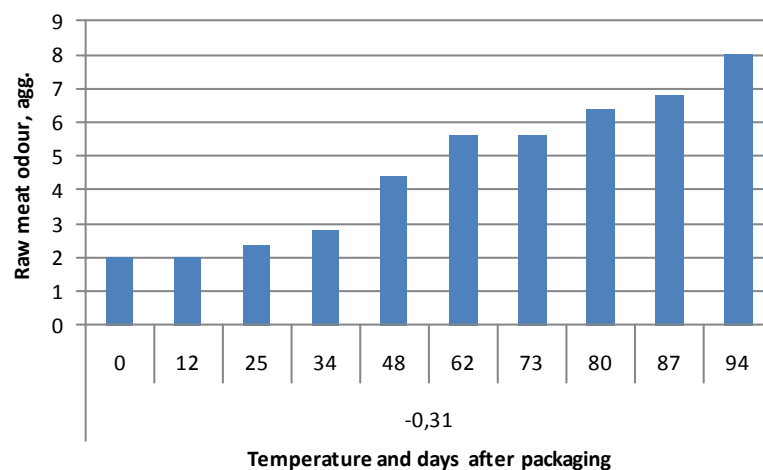


Figure 3: Development in raw meat odour during storage at -0.31°C . Bars show average values; $n=5$ per bar.

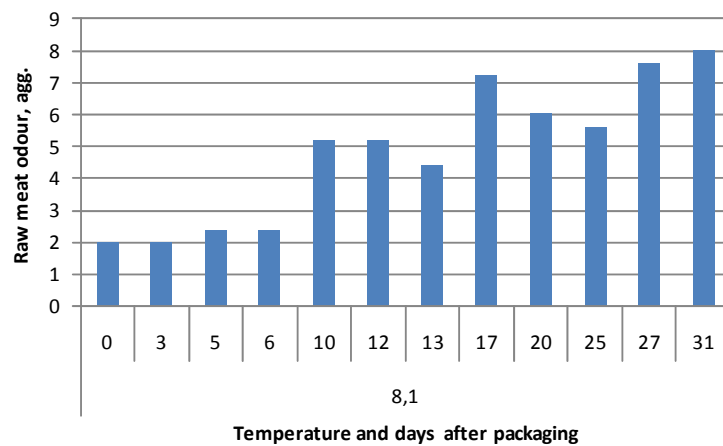


Figure 4: Development in raw meat odour during storage at 8.1 °C. Bars show average values; n=5 per bar.

In one package (Topside) the visual appearance was scored 4 and the raw odour 4. In 8 packages (Topsides, Sirloins and Cuvetter) the visual appearance was scored 3 and the raw meat odour 4. In all other 296 packages the raw meat odour was scored unacceptable before the visual appearance. The packages, for which visual appearance was scored unacceptable before the raw meat odour, were stored at both low and high temperatures and were without any correlation to gas composition.

Based on the above it is reasonable to use the raw meat odour for modelling of shelf life. But it should be noted that in a few percent of the packages, the visual appearance was unacceptable before the raw meat odour became unacceptable.

Changes in gas composition during storage

In the empty packages the content of O₂ and CO₂ was 20.0 % +/- 0.5 % and 81.9 % +/- 0.3 % (n=15 - 5 packages each time cuts were packed). The results indicate that the accuracy of the device used for measuring the gas composition, is within +/- 2 %, which is sufficient for this purpose.

Gas composition measured during storage at all temperatures is shown graphically in Annex 3.

During the first days of storage the % of CO₂ decreases to approx. 60 %, and the oxygen content increases. This is most probably due to some of the CO₂ dissolving in the meat.

The changes during storage depend on temperature. At -0.3 °C, after the initial drop in CO₂ concentration, there is a slight increase in CO₂ concentration and a slight decrease in O₂ concentration. At the three storage temperatures ≥ 3.4 °C, the O₂ concentration decreases towards 0 % and the CO₂ concentration increases towards 100 % during storage.

Spoilage appears as a result of intrinsic enzymatic activity and enzymatic activity arising from growth of micro-organisms. The fact that the changes in gas composition depends on temperature suggests that the part of the spoilage, which is caused by growth of micro-organisms decreases when the temperature approaches 0 °C, or that the flora, which causes the spoilage, differs with temperature.

pH - initial and during storage

Results from pH measurements at packaging are summarized in table 1.

At packaging, pH in Topsides differ significantly from pH in Cuvetter and Sirloins (p<0.002 - analysis of variance, SAS, proc GLM combined with Duncan's multiple range test).

pH seems to decrease during storage with the effect most pronounced at higher temperatures, see Figure 5.

Table 1: pH at packaging

	Cut		
	Cuvetter	Sirloin	Topside
Mean	5.59	5.57	5.65
Std.	0.06	0.05	0.08
Minimum	5.51	5.47	5.55
Maximum	5.73	5.67	5.80
N	15	15	15

Analysis of variance (SAS, proc GLM) shows significant effect ($p < 0.01$) of “day after packaging” on pH for all temperatures except 1.2 °C.

The decline in pH during storage could be caused by dissolved CO₂.

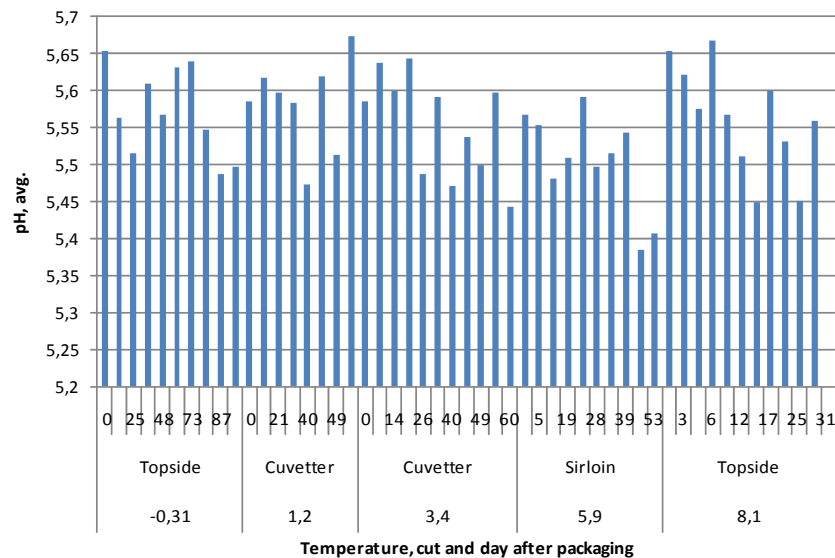


Figure 5: Development in pH during storage at all temperatures. Bars show average values; n=5 per bar with the exception of day 0, where n=15.

Growth of psychrotrophs during storage

Psychrotropic count during storage has been determined by two methods:

- Psychrotrophs - PCA - incubation at 6.5 °C for 10 days
- Psychrotrophs - IDF 132 (ISO 8552)

In the following, the two methods are referred to as “Milk, psky count” and “PCA, psky count”.

A detailed summary of results is found in Annex 4 and Annex 5.

As an example, the results from storage at 3.4 °C are shown in Figure 6.

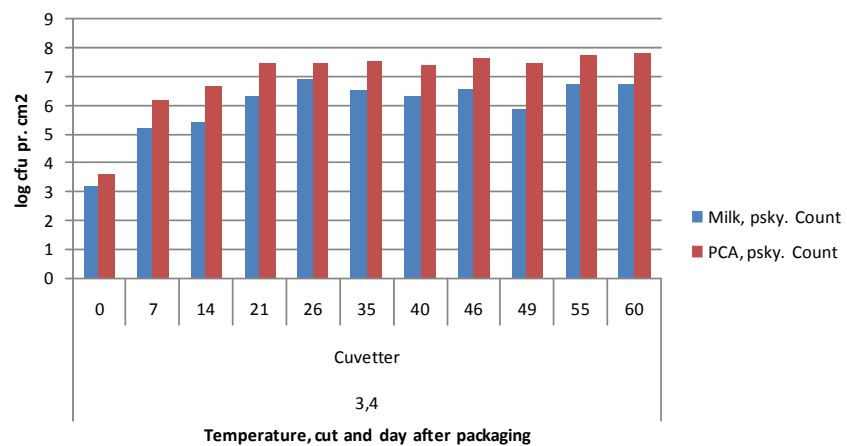


Figure 5: Psychrotrophic count during storage. log cfu per cm². Bars show average values; n=5 per bar with the exception of day 0, where n=15.

The maximum count differs between the two parameters and “between temperatures”.

At -0.31 °C, 1.2 °C and 3.4 °C (Topsides and Cuvetter) Milk, psky count reaches a maximum count between 6 and 7 log per cm² and PCA, psky count reach a maximum count between 7 and 8 log units. At 5.9 °C the max. count for both parameters is approx. 0.5 log unit lower and at 8.1 °C on topsides max count for both parameters is approx. 0.5 log unit higher.

The difference between the two methods is in average 0.63 log units and seems fairly constant.

It should be noted that determining the number of psychrotrophs according to IDF 132 is laborious and time consuming. A lot of the colonies are pin points, and as stated in IDF 132 it is necessary to use a magnifier when counting the colonies.

Modelling

When fitting the growth curves, N_{∞} was assigned the value of 6.5 and 7.5 log cfu per cm² for Milk, psky count and PCA, psky count.

Constants found, when fitting the growth curves for Milk, psky count, are shown in Table 2. Constants found, when fitting the growth curves for PCA, psky count, are shown in Table 3.

Table 2: Constants for modelling the growth of Psychrotrophs (IDF 132 (ISO 8552)) in MAP packed cuts of beef. The constants have been found when fitting 5 growth curves covering temperatures ranging from -0.31 °C to 8.1 °C. RMSE=0.775

	Estimate	Approx. Std. Error	Approx. 95% Confidence limits	
β_0	0.00635	0.0021	0.00212	0.0106
β_1	0.1963	0.0388	0.1179	0.2746
h	-0.5011	1.3046	-3.1338	2.1316

Table 3: Constants for modelling the growth of Psychrotrophs (PCA - incubation at 6.5 °C for 10 days) in MAP packed cuts of beef. The constants have been found when fitting 5 growth curves covering temperatures ranging from -0.31 °C to 8.1 °C. RMSE=0.779

	Estimate	Approx. Std. Error	Approx. 95% Confidence limits	
β_0	0.00606	0.00163	0.00276	0.00935
β_1	0.1933	0.0303	0.1322	0.2544
h	-1.6602	1.1424	-3.9657	0.6454

The contents describing the growth of Milk, psky count and PCA, psky count are remarkably alike indicating that it is the same flora, which is determined by the two methods.

For both counts the constant determined with the biggest uncertainty is h , which determines the lag-phase.

It is most unlikely, that h is below 0. In that case growth is faster just after packing than during the exponential growth phase. Therefore h is assumed to be 0 and the growth curves have been fitted with h assigned the value 0 before fitting - results are in Table 4 and Table 5.

Table 4: Constants for modelling the growth of Psychrotrophs (IDF 132 (ISO 8552)) in MAP packed cuts of beef. The constants have been found when fitting 5 growth curves covering temperatures ranging from -0.31 °C to 8.1 °C and with h assigned the value 0. RMSE=0.774

	Estimate	Approx. Std. Error	Approx. 95% Confidence limits	
β_0	0.00701	0.000125	0.00449	0.00953
β_1	0.1990	0.0365	0.1254	0.2725
h	0	-	-	-

Table 5: Constants for modelling the growth of Psychrotrophs (PCA - incubation at 6.5 °C for 10 days) in MAP packed cuts of beef. The constants have been found when fitting 5 growth curves covering temperatures ranging from -0.31 °C to 8.1 °C and with h assigned the value 0. RMSE=0.779

	Estimate	Approx. Std. Error	Approx. 95% Confidence limits	
β_0	0.00853	0.00117	0.00618	0.0109
β_1	0.2053	0.0284	0.1481	0.2625
h	0	-	-	-

Plots of found values against fitted curves, when using the constants in Table 4 and 5, are shown in Annex 6 and Annex 7.

Constants found, when fitting the curves for development in raw meat odour are shown in Table 6 and Table 7 - one for each method to determine the number of psychrotrophs, as this influences the count at packaging and thereby the "lag-phase".

Table 6: Constants for modelling the development of "Raw meat odour" in MAP packed cuts of beef. The constants have been found when fitting 5 growth curves covering temperatures ranging from -0.31 °C to 8.1 °C - NB: Psychrotrophs (IDF 132 (ISO 8552)). RMSE=1.205.

	Estimate	Approx. Std. Error	Approx. 95% Confidence limits	
β_0	0.00609	0.000949	0.00417	0.008
β_1	0.1265	0.0197	0.0867	0.1662
$\log(N_{lugt})$	2.9733	1.0981	0.7573	5.1893

Table 7: Constants for modelling the development of “Raw meat odour” in MAP packed cuts of beef. The constants have been found when fitting 5 growth curves covering temperatures ranging from -0.31 °C to 8.1 °C - NB: Psychrotrophs on PCA - incubation at 6.5 °C for 10 days. RMSE=1.217

	Estimate	Approx. Std. Error	Approx. 95% Confidence limits	
β_0	0.00606	0.000938	0.00417	0.00795
β_1	0.1260	0.0195	0.0866	0.1654
$\log(N_{lugt})$	3.1476	1.0987	0.9304	5.3648

Also when fitting the curves for development of raw meat odour it is the constant determining the “lag-phase”, which is determined with the largest uncertainty.

As there is a systematic difference in the number of psychrotrophs depending on the method of enumeration, this also influences $\log(N_{lugt})$ and to a lesser degree the other two constants.

Found values versus fitted values are shown as plots in Annex 8 and 9.

The values for $\log(N_{lugt})$ in Table 5 and Table 6 gives a “lag-phase” which is close to 0. This is in contrast to the observed time before the first changes in raw meat odour appears. The time passing, before a change in the raw meat odour is observed, is 12 - 25 days at -0.31 °C, 11 - 21 days at 1.2 °C, 7 - 14 days at 3.4 °C, 5 - 12 days at 5.9 °C and 3 - 5 days at 8.1 °C. This corresponds to a count of 4 - 5 log cfu per cm² and 5 - 5.5 log cfu per cm² at onset of changes for Milk, psky count and PCA, psky count, respectively.

Due to the above curves, describing raw meat odour have been refitted with $\log(N_{lugt})$ given the values of 4.7 and 5.3 log cfu per cm² for Milk, psky count and PCA, psky count respectively. Results are shown in Table 8 and 9. Found values versus fitted values are shown as plots in Annex 10 and 11.

As can be seen in Annex 10 and 11, the constants in Table 8 and 9 gives a better prediction in the first half of the shelf life period than the constants in Table 6 and 7. Furthermore, the RMSE is reduced by using the constants in Table 8 and 9. Therefore the constants in Table 8 and 9 are used in the two spreadsheets containing the “interface” for the models.

Table 8: Constants for modelling the development of “Raw meat odour” in MAP packed cuts of beef. The constants have been found when fitting 5 growth curves covering temperatures ranging from -0.31 °C to 8.1 °C - NB: Psychrotrophs (IDF 132 (ISO 8552)). $\log(N_{lugt})$ assigned a value of 4.7. RMSE=1.174.

	Estimate	Approx. Std. Error	Approx. 95% Confidence limits	
β_0	0.00736	0.000620	0.00611	0.00861
β_1	0.1287	0.0176	0.0932	0.1641
$\log(N_{lugt})$	4.7	-	-	-

Table 9: Constants for modelling the development of “Raw meat odour” in MAP packed cuts of beef. The constants have been found when fitting 5 growth curves covering temperatures ranging from -0.31 °C to 8.1 °C - NB: Psychrotrophs on PCA - incubation at 6.5 °C for 10 days. $\log(N_{lugt})$ assigned a value of 5.3. RMSE=1.170.

	Estimate	Approx. Std. Error	Approx. 95% Confidence limits	
β_0	0.00763	0.000622	0.00638	0.00889
β_1	0.1296	0.0170	0.0953	0.1640
$\log(N_{lugt})$	5.3	-	-	-

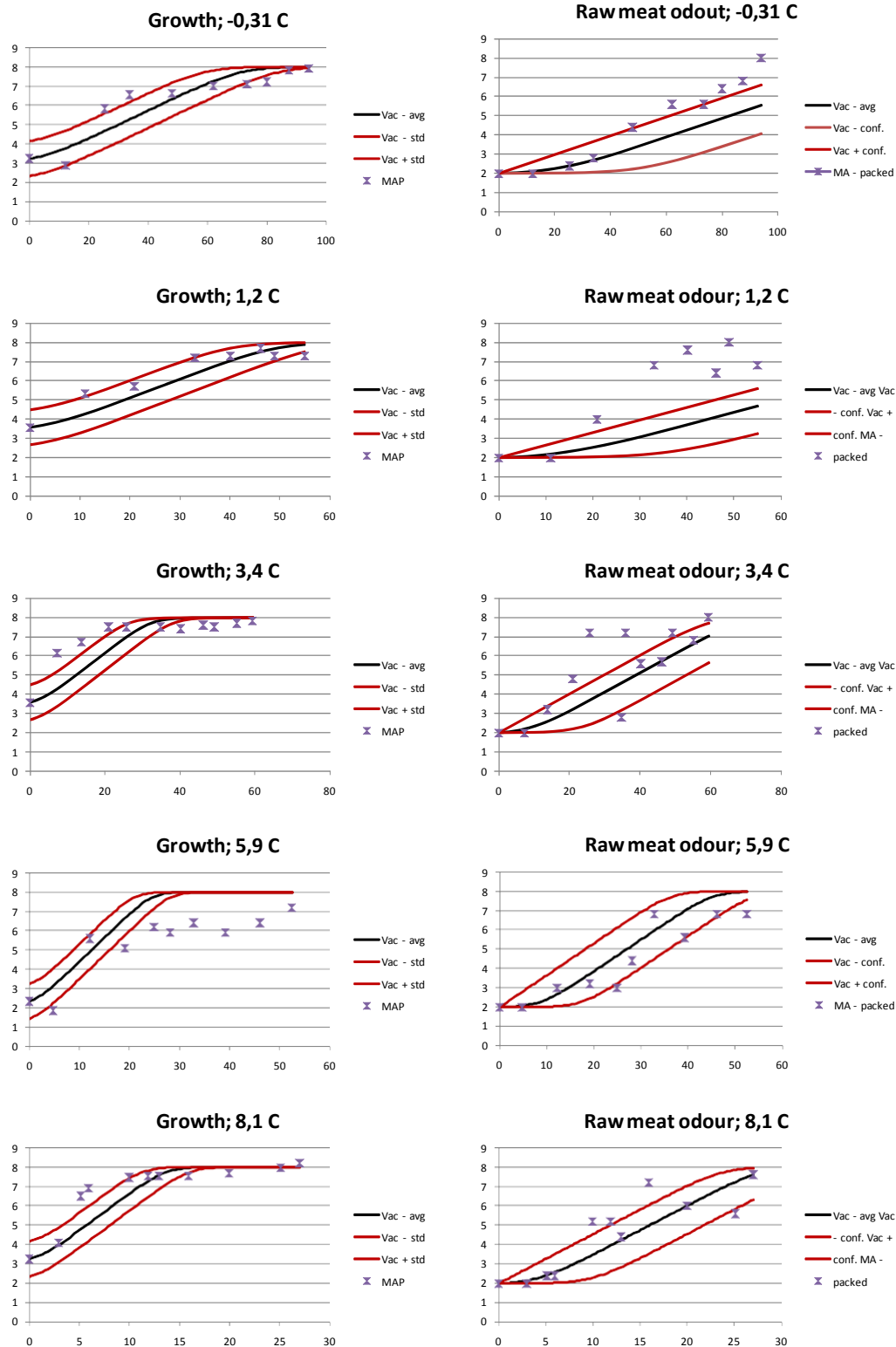
RMSE when comparing fitted and found values is approx. 0.8 and 1.2 for growth and development in raw meat odour, respectively. When developing the model for shelf life of vacuum packed beef, an RMSE of 0.38 and 0.78 were found for growth and raw meat odour. In a model recently developed for minced pork meat, the RMSE for growth and raw meat odour were 0.38 and 0.52.

The above implies that the differences between observed and fitted values are greater in the model for MAP packed beef than in the model for vacuum packed beef. In principle, the reason can be either larger variation in the raw material or that the packing method in itself “induces” the variation.

Shelf life of MAP packed beef compared with shelf life of vacuum packed beef

In Figure 6 growth (Psychrotrophs - PCA) and development of raw meat odour during MAP pack at all 5 temperatures are shown together with the expected growth and development of raw meat odour during vacuum pack at the same temperatures.

Figure 6: Growth (Psychrotrophs - PCA) and development of raw meat odour during MAP pack (80 % CO₂ + 20 % O₂) and expected growth and development of raw meat odour during vacuum pack at -0.31 °C, 1.2 °C, 3.4 °C, 5.9 °C and 8.1 °C.



The difference between counts found and scores for raw meat odour during storage of MAP packed beef is not very different from what would be expected in vacuum packed beef. There is a tendency that the difference is bigger at low temperatures than at high temperatures in the range of -0.3 - 8 °C.

Annex 1: Microbial profiles at the beginning of storage

		Log cfu per cm ²		
		Mean	Std	N
Cuvelter	Lactic acid bacteria	2.21	0.86	15
	Psychrotrophs - IDF 132	3.17	0.68	15
	Psychrotrophs - PCA	3.59	0.57	15
	Aerobic count	3.63	0.59	15
	Enterobacteriaceae	0.78	0.18	15
Sirloin	Lactic acid bacteria	1.00	0.37	15
	Psychrotrophs - IDF 132	1.75	0.58	15
	Psychrotrophs - PCA	2.36	0.74	15
	Aerobic count	2.49	0.62	15
	Enterobacteriaceae	0.74	0.11	15
Topside	Lactic acid bacteria	2.31	0.82	15
	Psychrotrophs - IDF 132	3.35	0.81	15
	Psychrotrophs - PCA	3.25	1.00	15
	Aerobic count	3.55	0.81	15
	Enterobacteriaceae	1.58	0.92	15

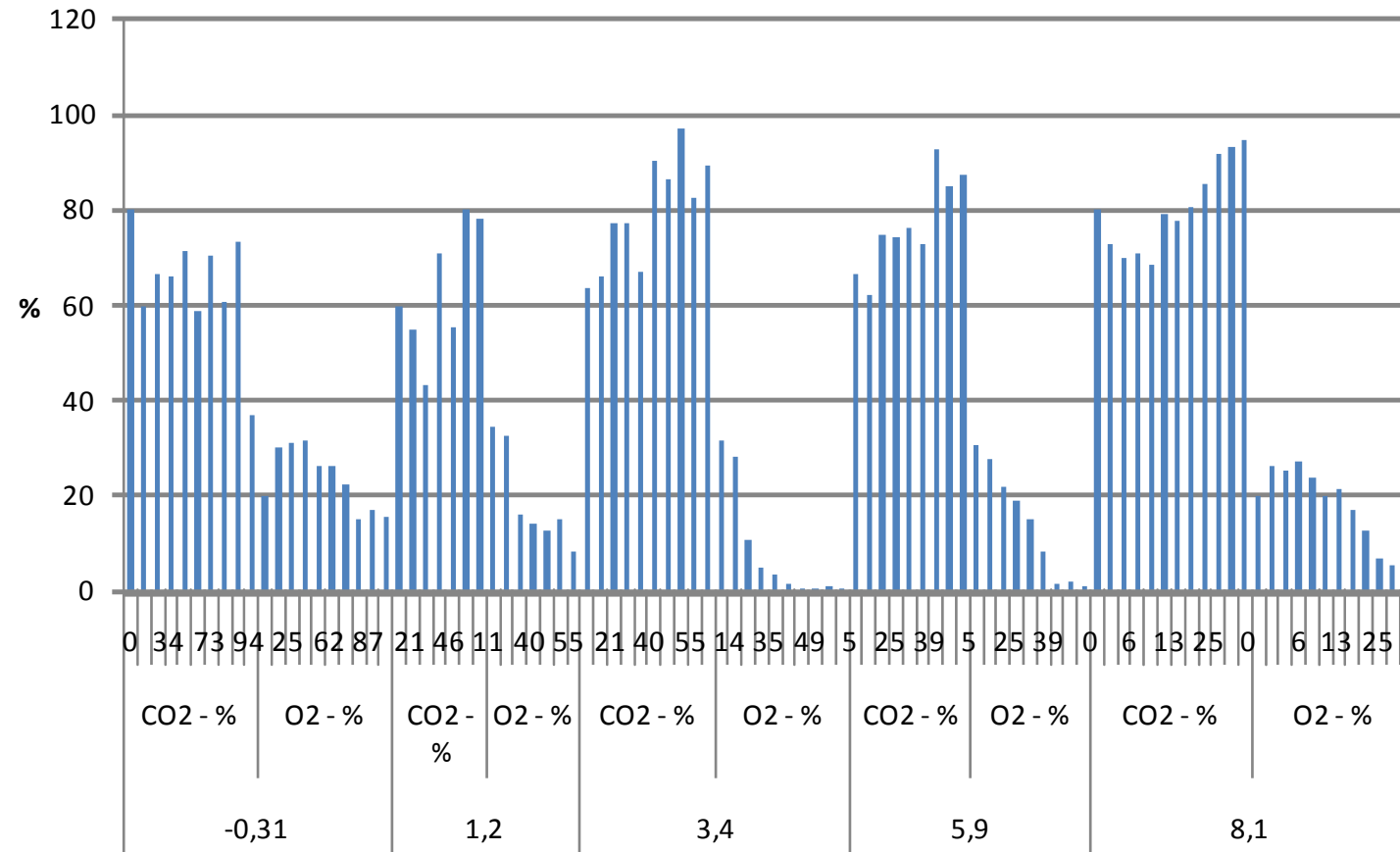
Annex 2. Development of raw meat odour during storage. Scale - 2: fresh; 4: slightly diverging but acceptable; 6: diverging to an unacceptable degree; 8: putrid/rotten.

Storage temperature, °C (cut)	Days after pack.	Raw meat odour		
		Mean	Std	n
-0.31 °C (Topside)	0	2	0	15
	12	2	0	5
	25	2.4	0.89	5
	34	2.8	1.10	5
	48	4.4	0.89	5
	62	5.6	0.89	5
	73	5.6	0.55	5
	80	5.4	0.89	5
	87	5.8	1.10	5
	94	8	0	5
1.2 °C (Cuvetter)	0	2	0	15
	11	2	0	5
	21	4	0	5
	33	6.8	1.1	5
	40	7.6	0.89	5
	46	6.4	0.89	5
	49	8	0	5
	55	6.8	1.10	5
3.4 °C (Cuvetter)	0	2	0	15
	7	2	0	5
	14	3.2	1.10	5
	21	4.8	1.10	5
	26	7.2	1.10	5
	35	2.8	1.79	5
	40	7.2	1.10	5
	46	5.6	0.89	5
	49	7.2	1.10	5
	55	6.8	1.10	5
	60	8	0	5

Annex 2 - continued. Development of raw meat odour during storage. Scale - 2: fresh; 4: slightly diverging but acceptable; 6: diverging to an unacceptable degree; 8: putrid/rotten.

Storage temperature, °C (cut)	Days after pack.	Raw meat odour		
		Mean	Std	n
5.9 °C (Sirloin)	0	2	0	15
	5	2	0	5
	12	3	0	5
	19	3.2	1.79	5
	25	3	0	5
	28	4.4	0.89	5
	33	6.8	1.10	5
	39	5.6	2.19	
	46	6.8	1.10	5
	53	6.8	1.10	5
8.1 °C (Topside)	0	2	0	15
	3	2	0	5
	5	2.4	0.89	5
	6	2.4	0.89	5
	10	5.2	1.10	5
	12	5.2	0.45	5
	13	4.4	0.89	5
	17	7.2	1.10	5
	20	6	0.00	5
	25	5.6	1.67	5
	27	7.6	0.89	5
	31	8	0.00	5

Annex 3 - Gas composition during storage. x-axis - day after packaging, gas, storage temperature.



Annex 4. Psychrotropic count (IDF 132) during storage

Storage temperature, °C (cut)	Days after pack.	log cfu per cm ²		
		Mean	Std	n
-0.31 °C (Topside)	0	3.35	0.81	15
	12	2.24	0.79	5
	25	5.81	0.43	5
	34	6.17	0.65	5
	48	6.15	0.81	5
	62	6.69	0.92	5
	73	6.43	0.57	5
	80	6.43	0.60	5
	87	7.03	0.66	5
	94	7.36	0.30	5
1.2 °C (Cuvetter)	0	3.17	0.68	15
	11	4.74	1.56	5
	21	4.65	0.96	5
	33	5.67	0.51	5
	40	6.31	0.27	5
	46	7.67	0.67	5
	49	6.29	0.17	5
	55	6.04	0.27	5
3.4 °C (Cuvetter)	0	3.17	0.68	15
	7	5.21	0.63	5
	14	5.41	0.61	5
	21	6.34	0.99	5
	26	6.93	0.47	5
	35	6.50	0.12	5
	40	6.30	0.65	5
	46	6.57	0.11	5
	49	5.84	0.33	5
	55	6.74	0.25	5
	60	6.74	0.56	5

Annex 4 - continued. Psychrotropic count (IDF 132) during storage

Storage temperature, °C (cut)	Days after pack.	log cfu per cm ²		
		Mean	Std	n
5.9 °C (Sirloin)	0	1.75	0.58	15
	5	1.50	1.10	5
	12	5.02	1.18	5
	19	4.05	0.97	5
	25	5.87	0.70	5
	28	5.89	1.37	5
	33	5.68	1.13	5
	39	5.74	0.73	5
	46	5.62	0.58	5
	53	6.41	0.45	5
8.1 °C (Topside)	0	3.35	0.81	15
	3	3.92	0.95	5
	5	6.23	1.14	5
	6	6.44	0.66	5
	10	6.83	0.49	5
	12	7.01	0.46	5
	13	6.96	0.44	5
	17	7.24	0.74	5
	20	7.59	0.41	5
	25	7.47	0.61	5
	27	7.95	0.30	5

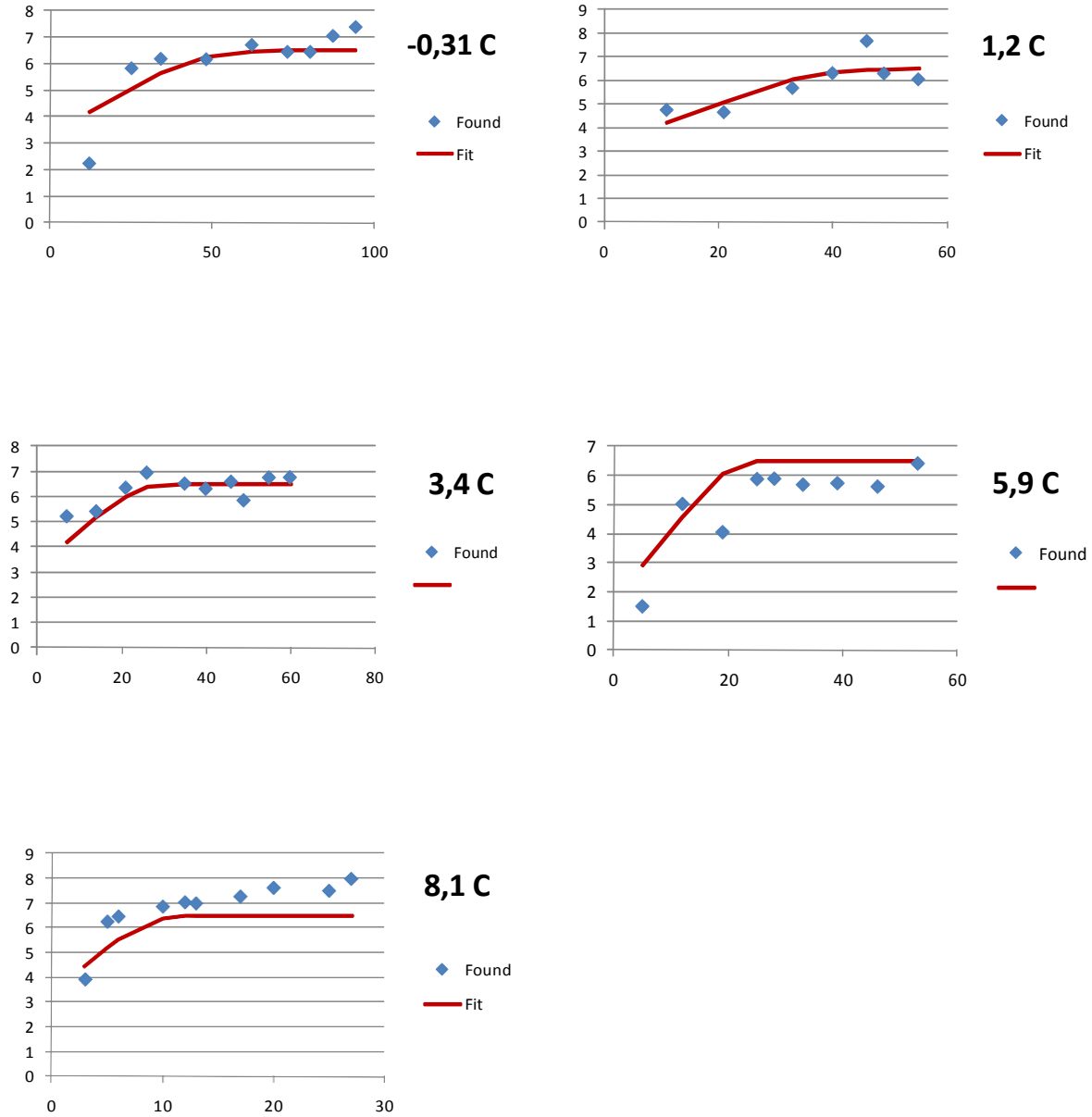
Annex 5. Psychrotropic count (PCA at 6.5 °C for 10 days) during storage

Storage temperature, °C (cut)	Days after pack.	log cfu per cm ²		
		Mean	Std	n
-0.31 °C (Topside)	0	3.25	1.00	15
	12	2.89	0.68	5
	25	5.83	0.61	5
	34	6.54	0.73	5
	48	6.62	0.79	5
	62	6.99	1.21	5
	73	7.09	0.65	5
	80	7.22	0.60	5
	87	7.83	0.50	5
	94	7.86	0.43	5
1.2 °C (Cuvetter)	0	3.59	0.57	15
	11	5.34	0.80	5
	21	5.69	0.92	5
	33	7.23	0.27	5
	40	7.32	0.20	5
	46	7.67	0.67	5
	49	7.34	0.14	5
	55	7.32	0.40	5
3.4 °C (Cuvetter)	0	3.59	0.57	15
	7	6.14	0.64	5
	14	6.67	0.73	5
	21	7.46	0.38	5
	26	7.48	0.38	5
	35	7.50	0.21	5
	40	7.40	0.15	5
	46	7.62	0.14	5
	49	7.45	0.28	5
	55	7.70	0.05	5
	60	7.83	0.21	5

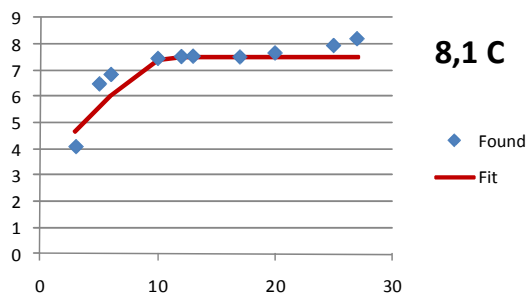
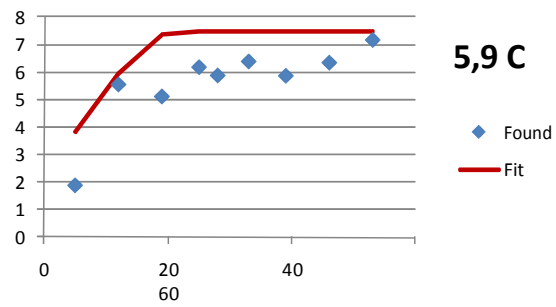
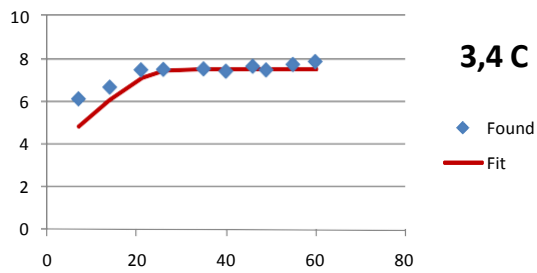
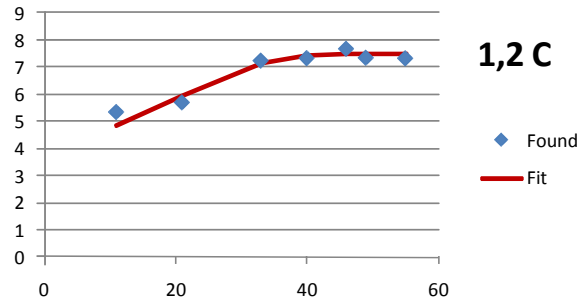
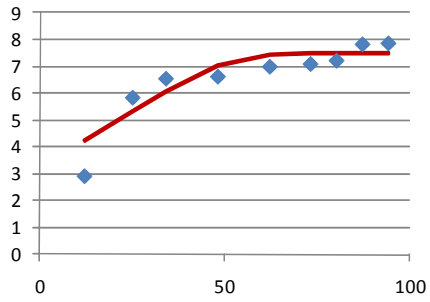
Annex 5 - continued. Psychrotropic count (PCA at 6.5 °C for 10 days) during storage

Storage temperature, °C (cut)	Days after pack.	log cfu per cm ²		
		Mean	Std	n
5.9 °C (Sirloin)	0	2.36	0.74	15
	5	1.88	1.18	5
	12	5.56	1.02	5
	19	5.12	0.94	5
	25	6.19	0.84	5
	28	5.89	4.41	5
	33	6.40	1.43	5
	39	5.87	1.60	5
	46	6.35	0.84	5
	53	7.18	0.72	5
8.1 °C (Topside)	0	3.25	1.00	15
	3	4.09	0.74	5
	5	6.49	1.05	5
	6	6.84	0.5	5
	10	7.46	0.48	5
	12	7.53	0.39	5
	13	7.54	0.23	5
	17	7.51	0.72	5
	20	7.66	0.64	5
	25	7.95	0.37	5
	27	8.21	0.34	5

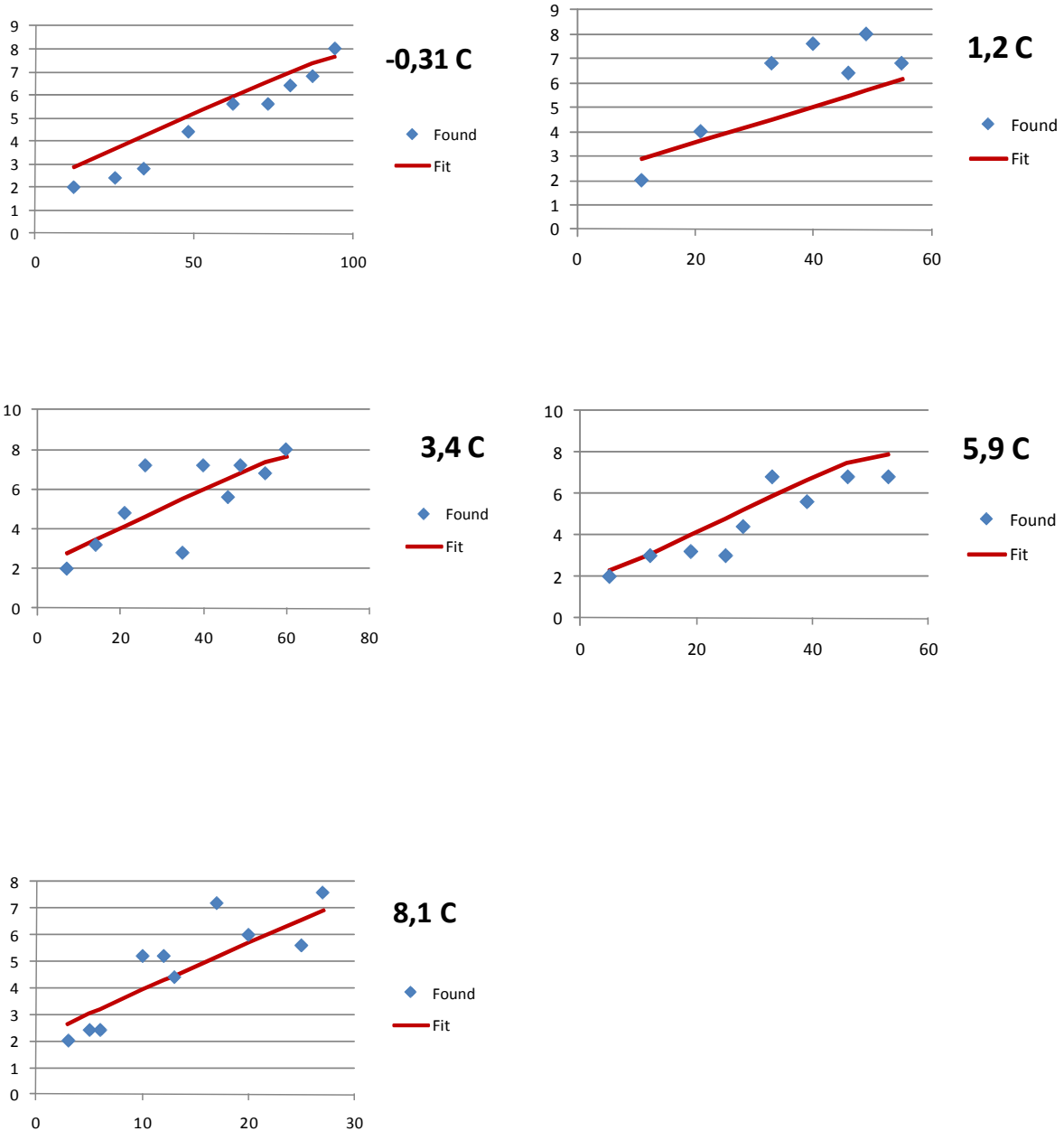
Annex 6: Fitted values versus found values for count - Psychrotrophs (IDF 132 (ISO 8552))
 $\beta_0=0.00701$. $\beta_1=0.1990$. $h=0$. Plot shows log cfu per cm² plotted against day after packaging at 5 storage temperatures



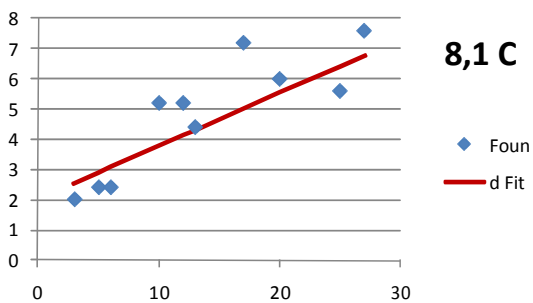
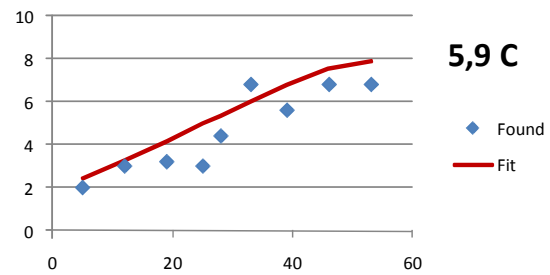
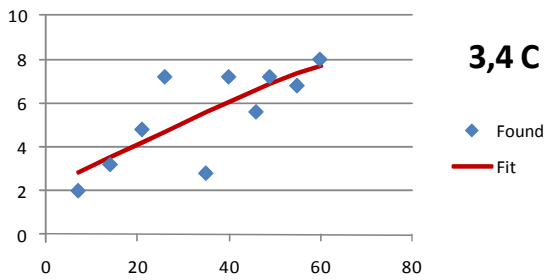
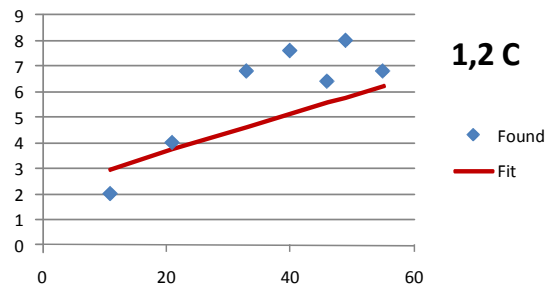
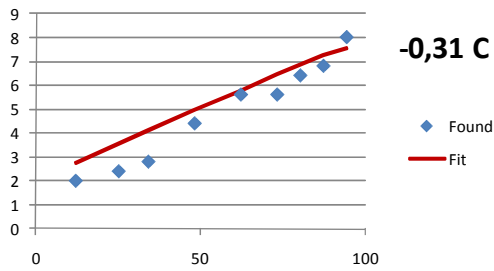
Annex 7: Fitted values versus found values for count - Psychrotrophs - PCA - incubation at 6.5 °C for 10 days. $\beta_0=0.00853$. $\beta_1=0.2053$. $h=0$. Plot shows log cfu per cm² plotted against day after packaging at 5 storage temperatures



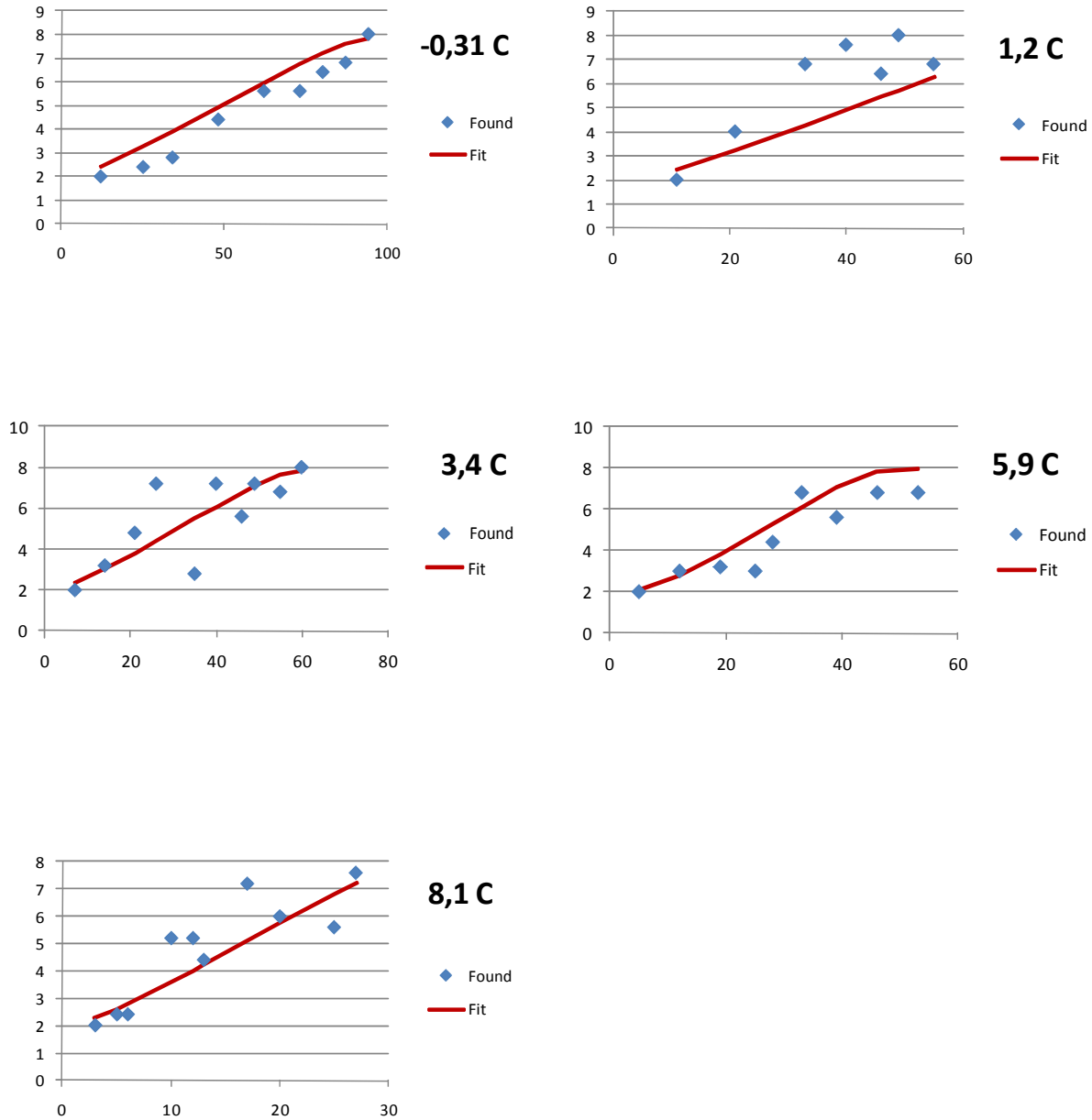
Annex 8: Fitted values versus found values for raw meat odour using the number of Psychrotrophs (IDF 132 (ISO 8552)) at day 0 to determine h . $\beta_0=0.00609$. $\beta_1=0.1265$. $\log(N_{lugt})=2.97$. Plot shows average score for raw meat odour plotted against day after packaging at 5 storage temperatures



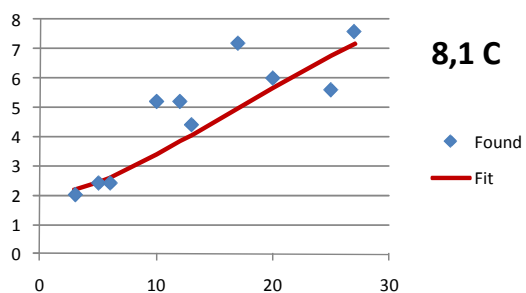
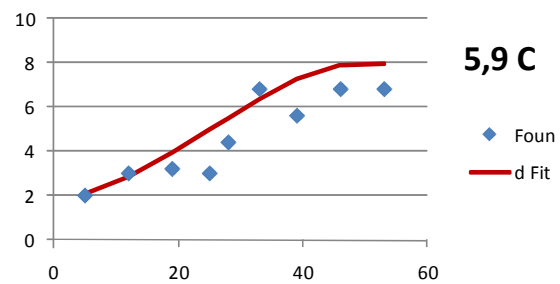
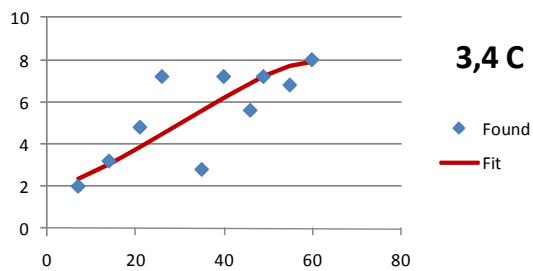
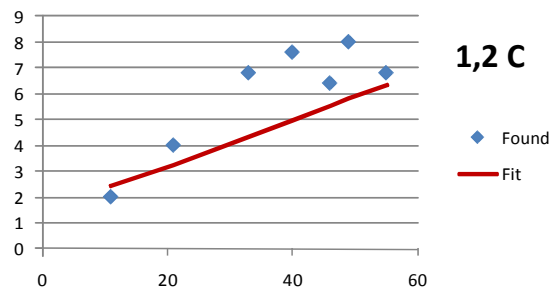
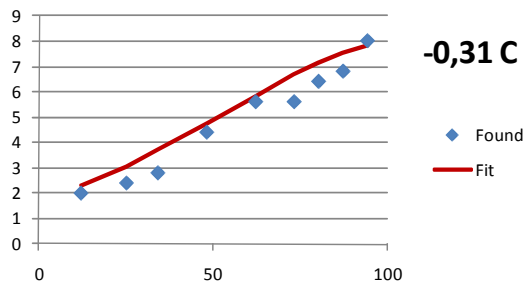
Annex 9: Fitted values versus found values for raw meat odour using the number of Psychrotrophs (PCA - incubation at 6.5 °C for 10 days) at day 0 to determine h . $\beta_0=0.00606$. $\beta_1=0.1260$. $\log(N_{lugt})=3.14$. Plot shows average score for raw meat odour plotted against day after packaging at 5 storage temperatures



Annex 10: Fitted values versus found values for raw meat odour using the number of Psychrotrophs (IDF 132 (ISO 8552)) at day 0 to determine h . $\beta_0=0.00736$. $\beta_1=0.1287$. $\log(N_{lugt})=4.7$. Plot shows average score for raw meat odour plotted against day after packaging at 5 storage temperatures



Annex 11: Fitted values versus found values for raw meat odour using the number of Psychrotrophs (PCA - incubation at 6.5 °C for 10 days) at day 0 to determine h . $\beta_0=0.00763$. $\beta_1=0.1296$. $\log(N_{lugt})=5.3$. Plot shows average score for raw meat odour plotted against day after packaging at 5 storage temperatures



Annex 12

TECHNICAL DATA SHEET

TOPSEAL ® PET MAP P B 62

For PET-trays.

TOPSEAL ® PET MAP P B 62 is a laminated barrier film, designed to give a medium barrier against O₂ and CO₂.

The film is peelable.

General property	Test Method	Unit	Value
Film Thickness (± 10%)		µm	62
Application Temperature		°C	+40 - +40
Sealing Temperature Range		°C	115 - 200
Area Yield (± 10%)		m ² /kg	15,4
Unit Weight (± 10%)		g/m ²	65,0
Core Diameter		mm	76
Permeability:			
- Oxygen	ASTM D3985, 23°C, 0%RH	cm ³ /m ² /24h/atm	<3,0
- WVTR	ASTM F1249, 38°C, 90%RH	g/m ² /24h	<1,5

Mechanical Property	Test Method	Unit	Value
Tensile strength at break - MD	ASTM D882	MPa	110
Tensile strength at break - TD	ASTM D882	MPa	120
Elongation - MD	ASTM D882	%	170
Elongation - TD	ASTM D882	%	200

Optical Property	Test Method	Unit	Value
Total Luminous Transmission	ASTM D1003-52	%	
Haze	ASTM D1003-52	%	

TOPSEAL ® PET MAP P B 62 is compliant with EC-directive 2002/72.

NormPack-certificate is available.