71st ICoMST
GIRONA - CATALONIA - SPAIN
3rd-8th August - 2025

PSEUDOMONAS IN MEAT: AN OLD LEGEND REVISITED

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I. INTRODUCTION

Due to its high nutrient and water content, meat is a highly perishable food commodity. The control of microbial spoilage largely depends on the effectiveness and maintenance of the cold chain. Under refrigeration conditions, psychrotrophic organisms prevail [1]. Amongst these, *Pseudomonas* is recognized as a spoiling agent in meat stored under aerobic conditions. *Pseudomonas* species associated with meat include notably *fragi*, *fluorescens* and *putida.* However, the opportunistic pathogen *aeruginosa* has raised concerns regarding meat safety. Poursina et al. [2] reported a prevalence rate of *P. aeruginosa* of 6.57 and 12% on meat and carcass surfaces, respectively. Additionally, their study reported antibiotic resistance amongst the isolates, with resistance levels ranging from 27.65 to 87.23% depending on the specific antibiotics tested.

According to the early work of Mead and Adams [3], Cetrimide, Fusidate and Cephaloridine (CFC supplement) are used to render the medium selective for *Pseudomonas* spp. in chilled foods, including meats. Initially, Heat Infusion (HI) served as the agar base [4]. Since then, *Pseudomonas* agar base has been developed and is now recommended for *P. aeruginosa* isolation. As per ISO 13720, this agar base is recommended for the enumeration of *Pseudomonas* spp. from meat and meat products. However, the question of whether the population of *Pseudomonas* spp. might be underestimated in meat when using *Pseudomonas* agar base, specifically designed for *P. aeruginosa*, warrants further investigation. Hence, our hypothesis suggests that an agar base with a composition closer to meat (Heat or Brain Heart infusion) could yield higher *Pseudomonas* spp. counts in meat products compared to the *Pseudomonas* agar base containing digest of gelatin and casein.

II. MATERIALS AND METHODS

A total of 28 meat samples (fresh or processed) from different animal species were collected in different local grocery and butcher stores. Samples were kept on ice until microbial enumerations were performed using CFC supplement with BHI, HI and *Pseudomonas* agar as base medium. Plates were incubated at 25°C for 48 h.

Statistical analysis was performed using R software (version 4.5.0) for the analysis of variance. When normality was not met (Shapiro-Wilk test), the Kruskal-Wallis test was used for non-parametric analysis by rank. Differences were considered significant when the *P*-value was lower than 0.05.

III. RESULTS AND DISCUSSION

Enumerations by animal species across the three medium agar bases are presented in Figure 1. No significant differences were observed between medium agar bases irrespective of whether the analysis included all samples ($P = 0.96$), only animal species (for beef, pork and rabbit, *P*-value varied from 0.93 to 0.98), fresh ($P = 0.93$) or processed meats ($P = 0.81$). Only the median rank indicates that BHI more frequently obtained the highest cell counts. When shelf-life regulations are determined by reaching a specific threshold, the medium providing the highest cell counts is more likely to attain that threshold more rapidly.

Similar experiments were carried out on fresh and frozen Black Soldier Fly larvae (*Hermetia illucens;* data not shown). In this instance, the median rank indicated that cell counts from *Pseudomonas* agar base exceeded those from BHI and HI, suggesting a higher level of contamination with *P. aeruginosa.*

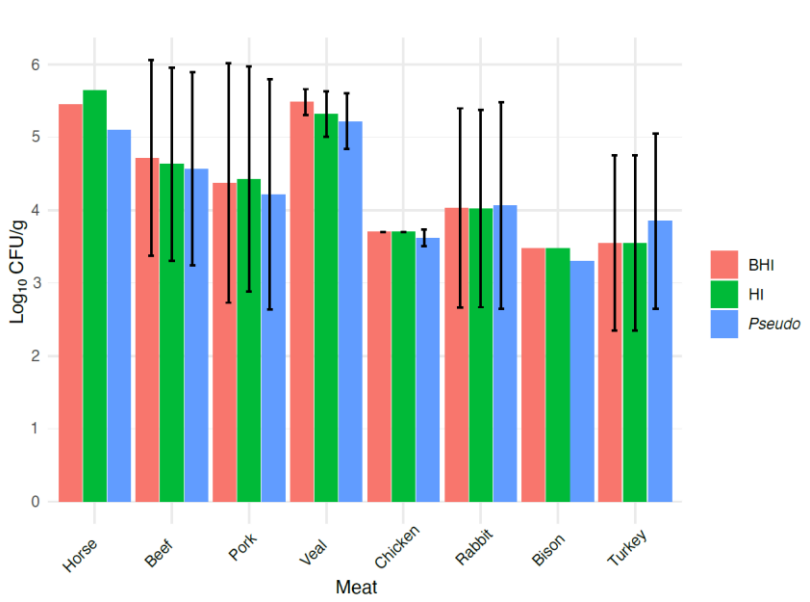


Figure 1. Enumeration of *Pseudomonas* spp. on meat using selective medium (CFC) prepared with three different medium agar bases.

Table 1 – Mean, standard deviation, total and median rank of *Pseudomonas* spp. enumeration using different medium agar bases.

	BHI	HI	*Pseudomonas* agar	*P*-value
All samples				
Means (log CFU/g)	4.35	4.34	4.26	0.96
Standard deviation	1.32	1.27	1.27	
Sum of ranks[#]	41	44	49	
Median rank	1	1	2	
Fresh meat				
Means (log CFU/g)	4.56	4.49	4.44	0.93
Standard deviation	1.14	1.10	1.08	
Sum of ranks	33	38	41	
Median rank	1	2	2	

[#] Ranks go from 1 to 3 where 1 was assigned to the highest cell counts.

IV. CONCLUSION

BHI as medium agar base presents a small advantage (median rank) when *Pseudomonas* spp. is used as spoilage indicator. However, to accurately evaluate the risk associated with *P. aeruginosa* in meat, its relative abundance in comparison to other *Pseudomonas* species should be examined using whole genome sequencing analysis throughout the storage period.

ACKNOWLEDGEMENTS

This research is part of the Canadian Food Innovation Network (CFIN; ASC-14) science cluster and funded in part by the Government of Canada under the Canadian Agricultural Partnership's AgriScience Program, which is a federal, provincial, and territorial initiative. The authors acknowledge the technical support of R. Courcoux.

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