

# Rapid Communication: A comparison of cardiac lesions and heart weights from market pigs that did and did not die during transport to one Ontario abattoir<sup>1</sup>

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**ABSTRACT:** In-transit losses of market hogs represent a small proportion of all market-weight pigs shipped in a year. This suggests that individual pig factors may be a significant cause of in-transit losses along with more traditionally considered environmental and transport factors. An investigation was performed to determine whether cardiac pathology and heart weights were associated with pigs that did or did not die during transport to an abattoir. The hearts from 70 pigs that died in-transit to one Ontario abattoir and 388 pigs that arrived alive were collected and examined. Hearts from pigs that died during transport demonstrated greater frequencies of cardiac lesions ( $P < 0.05$ ). These included hypertrophy of ventricle walls (Left: 97% vs. 64%; Right: 86% vs. 57%), dilation of ventricle

chambers (Left: 79% vs. 0.5%; Right: 100% vs. 5%), and dilation of the pulmonary artery and aorta (59% vs. 1.5%). Total heart weight to body weight ratios were increased (3.6 vs. 3.3 g/kg) and left ventricle plus septum weight over right ventricle weight ratio was decreased in pigs that died during transport over non-in-transit loss pigs (2.5 vs. 2.8;  $P < 0.05$ ). This may indicate reduced cardiac function in hogs that died during transport. Pigs with reduced cardiac function would have exercise intolerance and be more susceptible to death during transport due to the increased cardiac workload required during sorting, loading, and transport of the pigs to the abattoir. Further research to quantify cardiac function in pigs with cardiac lesions or abnormal heart weight ratios is warranted.

**Key words:** cardiac weights, cardiac ratios, in-transit loss, swine, ventricular hypertrophy, ventricular dilation

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## INTRODUCTION

In-transit losses (ITLs) of market weight pigs is defined as pigs that die during transport from the farm to the abattoir or pigs that become nonambulatory (NA) during transport and are euthanized on arrival to the abattoir (Ritter et al., 2009). ITLs in pigs are not fully explained by the commonly cited environmental risk factors such as temperature and stocking density and additional risk factors should be considered (Haley et al., 2008). The low proportion (0.7–0.8%) of market pigs that die during transport (Doonan et al., 2014) suggests that individual pig factors may be involved. Health problems of ITL pigs have not been well-studied and postmortem examinations at the abattoir are not routinely performed. In a preliminary study, the postmortem examinations of 85 ITL pigs from one Ontario abattoir indicated the cause of death to be heart failure (Zurbrigg et al., 2017). Pigs examined presented with pulmonary congestion and edema (62/85) and gross cardiac lesions of ventricular dilation, hypertrophy, and dilation of the major vessels (85/85). The researchers hypothesized that pigs with preexisting cardiac pathology were less likely to tolerate the physical exertion experienced during transport, which resulted in heart failure. However, hearts from pigs that did not die during transport should be examined for comparison purposes. This study compares the cardiac pathology and weights of hearts from ITL and non-in-transit loss (non-ITL) pigs.

## MATERIALS AND METHODS

### *Collection of Hearts From ITL Pigs*

The collection and examination of ITL and non-ITL hearts occurred simultaneously between June 2012 and April 2015. The ITL pigs and non-ITL pig hearts examined were collected from one federally inspected pig abattoir in Ontario, Canada which at the time received 15,000 to 20,000 pigs per week. Pigs were identified by a farm identifier (tattoo) which associated a pig with a geographic farm site and an owner. Farm site and owner information were known only to the abattoir staff.

The 70 ITL pigs examined were a convenience sample of all ITL pigs at the abattoir representing 40 loads. Carcasses were transported within 90 min of notification of the researcher, to the Animal Health Laboratory (AHL), University of Guelph, Ontario. Carcasses were weighed, body weight recorded (BW), and hearts were removed intact, rinsed, and stored in 10% formalin for future examination by one

pathologist who followed a standard protocol (see standardized heart examination protocol below).

An earlier study examined 85 carcasses and hearts from ITL pigs (Zurbrigg et al., 2017). The 70 ITL hearts in this paper were not part of that preliminary study.

### *Collection of Hearts From Non-ITL Pigs*

Hearts from non-ITL pigs were collected from the processing line at the same abattoir. Ten to 15 non-ITL hearts were selected from the processing line on each of 37 different days over the study period. The non-ITL pigs examined represent 74 loads. Hearts were selected by two different methods. The first method was to select the fifth non-ITL heart from each unique group of carcass tattoos as they came down the processing line until 10 hearts were collected (each from a unique tattoo) in 1 d ( $N = 100$ ). In the second method, hearts were collected from randomly selected farms shipping over 1,500 pigs per year which had given consent to the abattoir regarding participation for ITL studies. For these farms, hearts were collected from the processing line by selecting every  $n$ th heart, where  $n =$  the total number of pigs shipped from that farm identifier that day/10.

All the hearts from non-ITL pigs were removed from the processing line and placed in resealable freezer bags labeled with the farm identification number (tattoo) and the order in which the heart was collected (i.e., 1st–10th). Freezer bags containing hearts were placed in a cooler with ice packs and transported to the AHL. At the laboratory, the hearts were rinsed and placed intact into 10% formalin (within 1 h of collection) for future examination by one pathologist (T.v.D.) using the standardized protocol described below. Due to abattoir protocols, a specific BW for each non-ITL pig was not available. The mean BW and standard deviation for the load were provided and utilized as the BW for non-ITL hogs.

### *Standardized Heart Examination Protocol-ITL and Non-ITL Hearts*

A complete description of the standard protocol for examining the hearts is found in [Appendix 1 in Supplementary Material](#). The pathologist examining the hearts was blinded to whether the heart was designated as an ITL or non-ITL pig. After a minimum of 7 d of fixation in formalin, visible clots were removed from major vessels and noncardiac tissue and the aorta and pulmonary artery were trimmed

to their bases. The heart was then weighed and the weight recorded as total heart weight (TTHW). The aorta and pulmonary artery were observed for the presence of dilation of the lumen of the vessel and thinning of the vessel walls. The heart was then sliced transversely, approximately 1/3 of the distance between the base of the heart and the apex, and the presence of thickening of the chamber walls and dilation of the lumen of the left and right ventricles were recorded. Heart valves were examined and scored for the presence or absence of any thickening and/or nodularity. Hearts were sectioned into left ventricle and septum (LV + S) and right ventricles (RV) and each section weighed separately.

Four transmural sections of approximately 0.5-cm thickness and 1 cm<sup>2</sup> were taken perpendicular to the long axis of the left ventricle from each heart. They included the following: 1) ventricular septum (S) taken approximately one third of the distance between the aortic valve and the left ventricular apex; 2) posterior left ventricle (PLV) wall taken approximately half the distance between the mitral valve annulus and the left ventricular apex; 3) anterior left ventricle (ALV) wall sectioned approximately 2 cm lateral to the anterior descending coronary artery; and 4) right ventricle (RV) wall taken at the apex. In addition, a transverse section was taken of the proximal aorta dorsal to the aortic valves. Slides were prepared routinely and stained with hematoxylin-eosin. The pathologist recorded and described all lesions found in the sections.

### Statistical Analyses

The statistical program used for all analyses was Stata 14. Significance was set at  $P < 0.05$ . A Fishers exact test was used to determine whether there were significant differences in the frequencies of the gross and histologic cardiac lesions between hearts of ITL

and non-ITL pigs. The ratios of THW/BW and LVS/RV were calculated for each pig from the recorded TTHW, LVS, RV, and BW. A LVS/RV ratio of less than 2.8 was used as an indicator of RV hypertrophy (Robinson and Robinson, 2007). A  $t$ -test was used to determine whether there was a significant difference among the mean THW, LVS, RV, BW, THW/BW, and LV + S/RV of ITL and non-ITL pigs.

## RESULTS

### Heart Collection

The 70 hearts from ITL pigs came from 44 different farm sites. The 388 hearts from the non-ITL pigs came from 75 different farm sites.

### Gross Cardiac Lesions

Gross cardiac lesions were more frequently ( $P < 0.05$ ) observed in hearts from ITL pigs compared with non-ITL pigs (Table 1). Frequently hypertrophy and dilation were observed in the same heart. Combined dilation of the LV ventricle chamber and hypertrophy of the LV wall were observed in 77% (54/70) of ITL pig hearts. Combined dilation of the RV ventricle chamber and hypertrophy of the RV wall were observed in 85% (60/70) ITL pig hearts. Gross cardiac lesions were considered to have been present prior to transportation.

### Heart Weights and Body Weights

Heart weights, BWs, THW/BW, and LV + S/RV ratios for the hearts of ITL and non-ITL pigs are compared in Table 2. ITL pigs had greater TTHW, LV + S, RV, BW, and TTHW/BW ratios than non-ITL pigs ( $P < 0.05$ ). Non-ITL pigs had a greater LV + S/RV than ITL pigs ( $P < 0.05$ ) and the ratio was under 2.8 for ITL pigs.

**Table 1.** Gross cardiac lesions from the hearts of 70 ITL pigs and 388 non-ITL market pigs from one abattoir in Ontario, Canada

Gross lesion	ITL (%)	Non-ITL (%)
Thickened, nodular atrioventricular valves (endocardiosis)	35/70(50)*	14/388(3.6)
Hypertrophy of left ventricle wall	68/70(97)*	247/388(64)
Hypertrophy of right ventricle wall	60/70(86)*	221/388(57)
Dilation of chamber of left ventricle	55/70(79)*	2/388(0.5)
Dilation of the chamber of the right ventricle	70/70(100)*	21/388(5)
Dilation of the chambers of the atria (one or both)	47/70(67)*	2/388(0.5)
Dilation of the lumen of the aorta and pulmonary artery	41/70(59)*	6/388(1.5)
Thickened, fibrotic pericardial sac (pericarditis)	4/70(6)	1/388(0.25)

\* $P < 0.05$  for the comparison between the in-transit loss and non-in-transit loss values using a Fishers exact test.

**Table 2.** Heart weights of 70 ITL and 388 non-ITL market pigs from one abattoir in Ontario, Canada

Weight variable	ITL ( <i>SD</i> )	Non-ITL ( <i>SD</i> )
Total heart weight, g	464 (70.8)*	403.5 (49.9)
Left ventricle plus septum, g	282.6 (42.2)*	251.5 (29.2)
Right ventricle, g	113.4 (23.8)*	92.4 (14.2)
Left ventricle plus septum/ right ventricle [under 2.8 indicates RV hypertrophy]**	2.5 (0.33)*	2.8 (0.36)
Body weight, kg	128 (9.4)*	124.2 (5.7)
Total heart weight/body weight, g/kg	3.6 (0.52)*	3.3 (0.38)

\* $P < 0.05$  for the comparison of the mean ITL and non-ITL values using a *t*-test.

\*\*Robinson and Robinson, 2007.

### Histologic Lesions

The heart from one non-ITL pig and nine ITL pigs had one or more slides missing from the set and were not included in the histologic analyses. There were no differences ( $P > 0.05$ ), in the frequency of histologic lesions between hearts from 61 ITL and 387 non-ITL pigs with the exception of the greater cellular disarray and interstitial fibrosis observed in non-ITL hearts. The frequencies of histologic lesions for ITL vs. non-ITL pigs were as follows: medial hyperplasia of the intramural coronary arteries [53/61 (87%) vs. 332/387 (86%)], perivascular fibrosis of the intramural coronary arteries [47/61 (77%) vs. 308/387 (80%)], hypertrophy of the myocardial fibers [52/61 (85%) vs. 332/387 (86%)], atrophy and fatty replacement of myocardial fibers [13/61 (21%) vs. 80/387 (21%)], degeneration of myocardial fibers [3/61 (5%) vs. 14/387 (4%)], nuclear rowing (pleomorphism) of myocardial fibers [52/61 (85%) vs. 356/387 (92%)], cellular disarray of myocardial fibers [44/61 (72%) vs. 330/387 (85%);  $P < 0.05$ ], and interstitial fibrosis of myocardial fibers [4/60 (7%) vs. 79/387 (20%);  $P < 0.05$ ].

Appendix 2 in Supplementary Material contains photos demonstrating the gross lesions described and comparing an ITL heart with characteristic lesions to a non-ITL heart without lesions (Supplementary Figures 1–6).

## DISCUSSION

### Gross Cardiac Lesions

The frequency of LV hypertrophy, RV dilation of the ventricle walls, and dilation of the atria, aorta, and pulmonary artery in ITL pigs were similar to those found in a previous study involving a

postmortem examination of the entire carcass of pigs that died during transport (Zurbrigg et al., 2017). The observation of RV dilation in ITL hogs and the suggestion that this preexisting lesion resulted in acute heart failure of pigs during transport have been previously published (Bergmann, 1988; Zurbrigg et al., 2017). Hypertrophy of the ventricle walls and dilation in the lumen of the ventricles, aorta, and pulmonary artery indicate a heart in a state of compensatory or decompensatory heart failure (Lilly, 2011). Hypertrophy of the ventricle can turn from adaptive to maladaptive as the thickening ventricle wall begins to impair contractility and relaxation (Robinson and Robinson, 2007). Dilation of the LV chamber has been associated with electrical instability and death (Gaudron et al., 2001) and the RV chamber dilation has been associated with RV dysfunction and a poor prognosis in humans and pigs (Greyson et al., 2000; Madias et al., 2011). These conditions have also been associated with exercise intolerance in people (Pinsky 2016). During physical exertion, pigs may show signs of exercise intolerance through wheezing, trembling, and becoming NA. It is plausible that the significantly increased frequency of dilation and hypertrophy of the ventricles observed in ITL hearts over non-ITLs is associated with reduced tolerance to the exertion and stress of transport resulting in increased mortalities.

### Heart Weights and Body Weights

The THW, LVS, RV, THW/BW, and LVS/RV identified in this study were consistent with the weights observed in ITL hearts in a previous study (Zurbrigg et al., 2017). The heavier THW, LVS, RV and greater THW/BW ratio of ITL hearts is indicative of cardiac remodeling (Bienvenue et al., 1991; Carr et al., 2005). The lower LV + S/RV ratio (under 2.8) observed in ITL pigs has been associated with RV hypertrophy (Robinson and Robinson, 2007). Previous research has indicated increased THW/BW ratios in pigs with cardiac lesions observed at postmortem examination (Drolet et al., 1992; Liu et al., 1994; Huang et al., 2001).

The higher average BW of ITL pigs compared with non-ITL pigs implies an increased risk of transport death in heavier pigs. Studies by Rademacher and Davies (2005) and Nannoni et al. (2016) support this concept. It has been suggested that heavier pigs may show an increased metabolic response and temperature rise during transport compared with lighter weight pigs making them more susceptible to transportation losses (Ritter et al., 2009).



## Histologic Lesions

Some of the histologic lesions observed (e.g., medial hyperplasia and perivascular fibrosis of the intramural coronary arteries, hypertrophy and cellular disarray, nuclear enlargement, and nuclear rowing) are similar to those associated with hypertrophic cardiomyopathy (HCM) in humans (Hughes 2004) and an HCM-like condition in pigs (Liu et al., 1994; Shyu et al., 2002). However, there were no differences in the frequency of these lesions between ITL and non-ITL hearts. It is possible that a difference in the severity of the lesions was present but not captured with the present or absent scoring system. Additional studies are needed before histologic cardiac lesions should be used to differentiate between ITL and non-ITL hearts in swine.

## Limitations

Due to the abattoir protocol, a specific pig BW corresponding to each non-ITL heart could not be determined. However, the abattoir paid the highest prices for pigs that fell within a specific weight and back fat category. This encouraged low variation of BWs in a farm's shipments as was demonstrated by the low standard deviation of weights in a load (data not shown). Therefore, using the mean BW per shipment would have provided a close approximation of the actual BW of each pig.

## CONCLUSION

Examination of the hearts of 70 ITL pigs revealed gross cardiac lesions indicative of compensatory remodeling. The much greater frequency of these cardiac lesions in the hearts of ITL pigs compared with non-ITL pigs and the increased ratio of THW/BW and decreased ratio of LVS/RV of ITL pigs suggests that cardiac pathology is associated with ITLs. It is possible that pigs with decreased LVS/RV ratios (under 2.8) are unable to respond to the increased cardiac exertion required during sorting, loading, and transport to the abattoir which results in transport mortalities due to cardiac insufficiency. Further research is needed to determine what initiates cardiac remodeling in pigs and to measure cardiac function in pigs with lesions. A complete examination of the heart including total and sectioned cardiac weights and ratios should be a key component of a postmortem examination when determining the cause of death in ITLs.

## SUPPLEMENTARY DATA

Supplementary data are available at *Translational Animal Science* online.

*Conflict of interest statement.* None declared.

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