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Age-Dependent Material Characterization of Porcine Abdominal Organs

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ABSTRACT

The mechanical response of thoraco-abdominal solid organs subjected to compressive loads has been described in a limited number of studies and little is known about how this response fluctuates with age. The current study explores variations in the mechanical response of several porcine organs to small displacements over a wide range of ages. Seventeen female pigs ranging in age from 14 days to 211 days were used in this study. Based on necropsy analysis, this age range corresponds to a human abdominal development ranging from an infant to an adult. Each organ specimen was extracted and tested shortly after the animal was euthanized. A servomotor was used to apply 2 mm to 15 mm displacements with a 5.55 mm diameter spherical punch. A chamber was constructed to maintain an in vivo temperature environment while each organ specimen was subjected to a battery of displacements including non-injurious and injurious ramp-and-hold waveforms, as well as triangle waves over a frequency range of 0.25 Hz to 2.25 Hz. The force-penetration response of the liver, kidney, and lung is presented in this paper and evaluated for age dependence. Results from this study indicate a positive correlation between the tissue modulus and age for the porcine liver and kidney. The tissue modulus for the lung decreased with age. These results agree with past experimental findings in the literature. Future analyses will utilize quasi-linear viscoelastic theory and Boussinesq contact theory to generate constitutive models for each organ from the current study. The reduced relaxation functions and the instantaneous elastic models from these findings will be compared across ages to investigate fluctuations in the material response.

INTRODUCTION

The abdomen is the second most commonly injured body region in young children using adult seat belts. Injuries to this region are often a result of what is commonly known as “seat belt syndrome”. Children are at greater risk for abdominal belt loading than adults. Children’s Hospital of Philadelphia reports that 43% of children are sub-optimally restrained. This can be attributed to poor belt geometry with the shoulder belt under the occupant’s arm or behind the occupant’s back coupled with the smaller, more cartilaginous pelvis of a child that the lap belt can slide over more easily resulting in abdominal loading. Abdominal belt loading can lead to severe trauma for an occupant during a crash event. Trauma to the abdominal organs has

a high degree of mortality and potential for complications, and may result in extended hospitalization with high medical costs. Automotive design engineers, however, are limited in the tools available in their efforts to design restraint systems that mitigate abdominal injuries to children. Unfortunately, no current pediatric anthropometric dummy has the capability to quantify accurately the engineering response of the abdomen due to belt loading. Researchers need to understand this abdominal response to enhance the biofidelity of child dummies and to improve restraint designs. Studying the force-penetration characteristics of the abdominal organs and how these characteristics change during pediatric development is the first step towards understanding human pediatric abdominal response.

Many methods have been used to characterize the mechanical response of various organs such as the liver, lung, spleen, and kidneys (e.g., Hayes and Mockros, 1971; Lai-Fook et al., 1976; Seki and Iwamoto, 1998; Farshad et al., 1999; Tamura et al., 2002; Gefen et al., 2003). Some methods involve gross manipulation of the biological specimen during test preparation and these studies assume that such methods do not alter the response of the tissue. Other studies attempt to model the breaking tolerance or other injury criteria to establish an injury threshold. Injury characterization is not the goal of this research. This study investigates if there are differences in the material properties of intact solid organs for sub-injury displacements, and if these differences correlate with the age of the test specimen.

There have been several investigations of the material properties of soft tissues and organs and how these properties change with age. Vitek and Valenta (2001) found that arterial walls develop more collagen cross-linking with age, and fibroblasts produce more elastin and collagen fibers. This tends to “preload” the tissue, causing an increase in residual strain and stiffness. There have been several studies documenting the compliance of lung tissue as a function of age (e.g., Turner et al., 1968; Niewoehner et al., 1975; Bode et al., 1976; Colebatch et al., 1979; Mahler et al., 1986; Paleček and Ježová, 1988; Kurozumi et al., 1994; Lai-Fook and Hyatt, 2000), but these studies are typically focused on the mechanics of respiration and refer to the pressure-volume compliance of the lung during a uniform inflation, although they do support the notion of an increase in tissue modulus with age. This increase in modulus with age is consistent with the findings of Yeh et al. (2002) for the liver. They performed quasistatic compression tests of 19 fresh liver specimens and found that cirrhotic tissue, the presence of which was correlated with age, had greater elastic modulus than healthy tissue. Since liver fibrosis is known to increase with aging, it is not unreasonable to assume that the liver modulus generally increases with age. The increase in modulus from healthy to extremely fibrotic tissue was on the order of four times (approximately 1000 Pa to 4000 Pa).

Most studies, including the ones mentioned above, have investigated changes in the material properties of biological tissues for aging adults. There is actually little information describing material property changes in children and adolescents, especially for the soft tissues of the abdomen. It cannot be assumed that trends found in aging adults also apply to children because a child is in a state of biological development while similar tissues of aging adults may begin to deteriorate with age.

This research is a preliminary investigation of age-related changes in the material properties of pediatric porcine specimens. The methods used in this analysis focus on isolating changes in the mechanical response of the tissues from aberrations introduced by organ perfusion and pressurization, and from any pathologic anomalies caused by disease. This study focuses on age-related changes observed during pediatric development from infancy to early adulthood. A more robust study will be performed in the future, but the procedures and results presented in this paper provide the groundwork for further investigations and analysis.

METHODS

Pigs were chosen as the human surrogates for this study. Past studies have used swine as surrogates for humans in biological testing (e.g. Viano et al., 1989; Miller, 1989). Twelve female, Yorkshire *sus scrofa* from 6 different litters were used in this study. This group was mainly composed of pediatric and adolescent swine, but also included two adults. The age and mass of the swine when sacrificed ranged from 14 days to 211 days and from 3.75 kg to 101.2 kg. The litters of newborn female pigs were raised in a controlled environment and, once weaned, were put on a controlled diet. Once each test subject reached the appropriate age, it was anesthetized then euthanized using an appropriate dosage of pentobarbital. Following euthanization, the test subject was transported to the research lab and extensive anthropometric measurements were taken. Next, the subject was dissected and the organs were removed. The organs were

extracted within 2-3 hours of death and immediately measured to determine the volume and the mass. Using litters of pigs provided fresh (unfrozen/unembalmed), intact organ specimens for testing. A table summarizing each test subject's measurements and data is included in the appendix.

Organ Preparation

The goal of the organ preparation methodology was to maintain an *in situ* environment to reproduce accurate results and reduce variability in the responses. Care was taken to minimize error in the study that could be introduced through specimen preparation and testing. Organs were extracted shortly after death and immediately placed into bags of saline solution until they were ready to be tested. The bags of saline containing each organ were placed in a temperature bath that was set to 37°C (98°F) for approximately 30 to 45 minutes prior to testing. The organs were then removed from the bags of warm saline and placed into a sealed acrylic temperature chamber filled with saline solution at 37°C (98°F). If necessary, the organ was positioned using aluminum shims.

Test Fixture

This study utilizes indentation instead of other methods such as compression tests because indentation isolates the material response from the structural response. This is critical because the organs increase in size with age, which would have an effect on the organ structural response. A spherical punch was chosen as the interface between the linear actuator and the test specimen because the spherical shape avoids the problem of edge stress concentrations normally associated with cylindrical punches, and the solution to the punch contact problem is well developed (Lee and Radok, 1960; Sneddon, 1965; Hayes et al., 1972; Zeng et al., 1992; Yang, 2003). The spherical punch was 5.55 mm in diameter, composed of steel, had a mass of 10.5 grams, and was assumed to be rigid. The contact between the punch and the test specimen was assumed to be frictionless.

A custom test fixture was developed to apply various displacement waveforms to each organ specimen. The driving mechanism was an Industrial Devices Corporation custom-built linear actuator driven by a servomotor. The system was capable of displacements of up to 30 mm at velocities of up to 100 mm/s. The device was mounted vertically on a set of machinist's chucks, which allowed the device to be positioned so that the displacements were normal relative to the organ surface. Prior to each test, the organ specimen was preloaded to 0.005 N to ensure that the spherical punch was in contact with the organ's surface. Two displacement waveforms were applied to each organ after preloading. First, a precondition waveform consisting of a 10 cycle triangle wave was applied to the organ to produce a repeatable force response. A ramp-and-hold displacement was applied immediately following the precondition waveform. This waveform was used to compare the force response of each organ for a given displacement. For the comparison reported in this paper, 5 mm ramp-and-hold displacements were applied to each organ and the force at 3 mm of displacement was compared for all organs to assess the effect of age. Figure 1 shows an illustration of the test setup alongside a photograph of an organ displacement test.

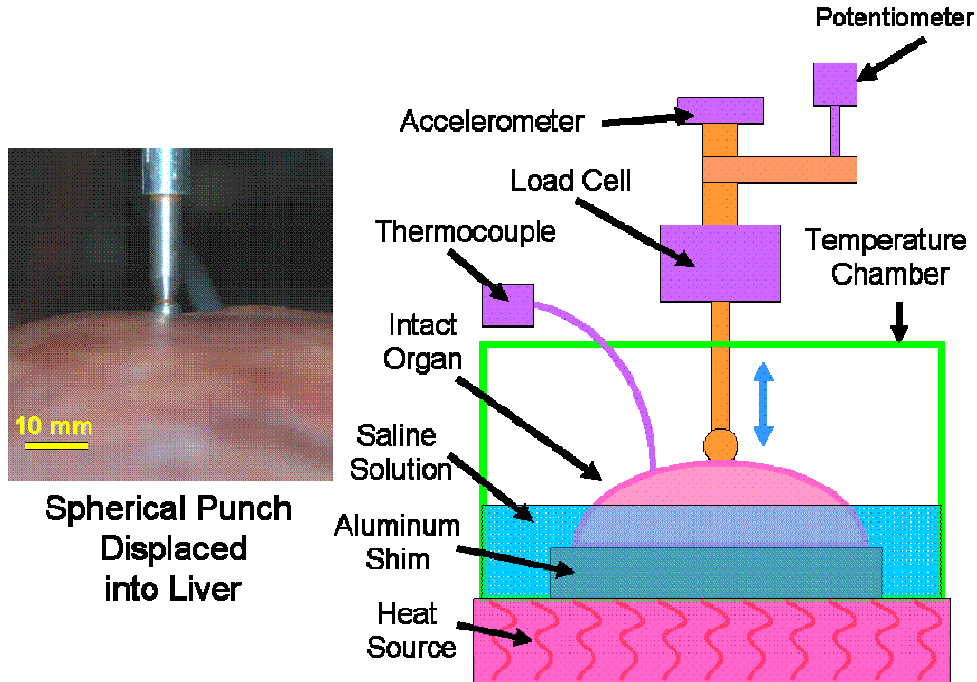


Figure 1: Photograph of Liver Displacement and Illustration of Test Setup.

Several sensors were used to measure the parameters necessary to study changes in the material properties of organs (Figure 1). A 100-g accelerometer was mounted to the linear actuator and measured accelerations of the punch in the direction of the punch displacement. A 100-gram uniaxial strain gage load cell was mounted between the linear actuator and the spherical punch and measured the load applied to the organ by the punch. A linear potentiometer was mounted on the linear actuator and measured the depth of punch penetration into the test specimen. The temperature of the organ surface was monitored using a thermocouple and the thickness (depth) of the organ was measured for each test.

Data Analysis

A series of comparisons for each organ was made among the force responses to 5 mm deflections. The first comparison was made to ensure that the force response of the organ did not depend on the location of the punch deflection. Three different locations were tested on the same organ specimen, with substantial space between locations so that any changes in the tissue due to a deflection would not be imposed on the next test. Another analysis compared the force response of two organs of nearly the same age, validating the test fixture's applicability across test subjects. If the differences between subjects of similar age were negligible, then response variations for subjects of different ages could be attributed to changes in age. Error (variation) was calculated by taking the standard deviation of the response as a percentage of the mean response.

To investigate changes in the material response with age, the force response for each organ was measured at 3 mm of deflection to yield an effective stiffness, K_{eff} . These values were then plotted against the age of the swine. This procedure results in a series of points whose magnitudes could be statistically evaluated for age correlation. A linear model was fit to the cross-plot of K_{eff} vs. swine age using a least-squares approach. The linear model produced a measure of K_{eff} for each organ as a function of test specimen age. The equation for the stiffness-age relationship is given below:

$$K_{\text{eff}} = \alpha + \beta \cdot \text{Age} \quad [1]$$

The significance of this model (Equation 1) was assessed by testing the null hypothesis, $H_0: \beta = 0$. The linear model provides a method to investigate changes in the effective stiffness of the organ as a function of age. The p-values, R^2 values, and the linear models are presented in the Results section.

RESULTS

Thirty-eight tests were performed successfully and used to develop age-dependent trends for changes in the effective stiffness of pediatric porcine thoraco-abdominal organs. In all tests, reasonable deflections were obtained and reasonable forces were measured. Figure 2 is a force-penetration cross plot for responses at three locations on the same kidney of a 132 day-old swine. Similar cross plots are shown for the liver of a 76 day-old swine (Figure 3) and for the lung of an 82 day-old swine (Figure 4). The three responses for the kidney are similar with a mean force of 0.2110 N (3 mm of deflection) and a standard deviation of 0.0042 N (2% error). The mean force was 0.1892 N (3 mm of deflection) with a standard deviation of 0.0182 N (10% error) for the liver. The mean force was 0.0730 N (3 mm of deflection) with a standard deviation of 0.0096 N (13% error) for the lung.

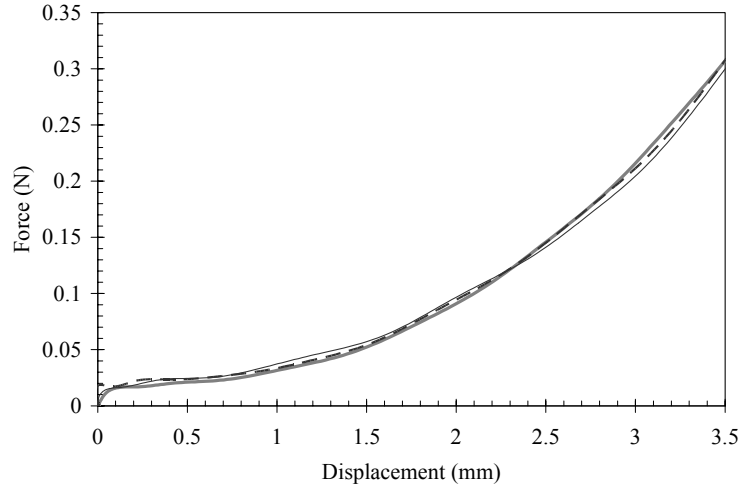


Figure 2: Force Response Variation by Location for Kidney Deflection.

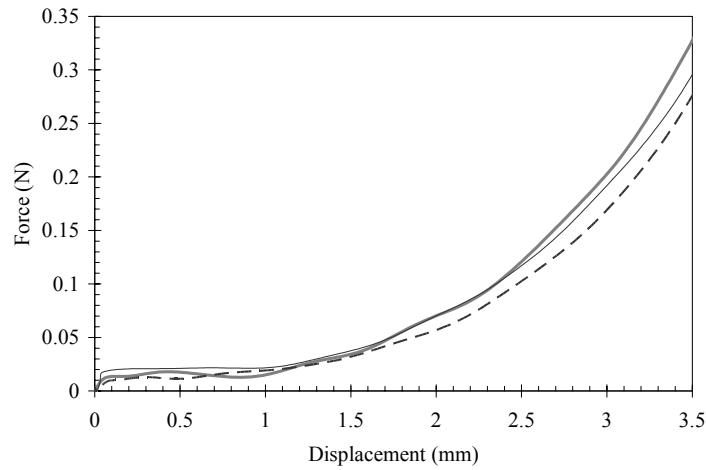


Figure 3: Force Response Variation by Location for Liver Deflection.

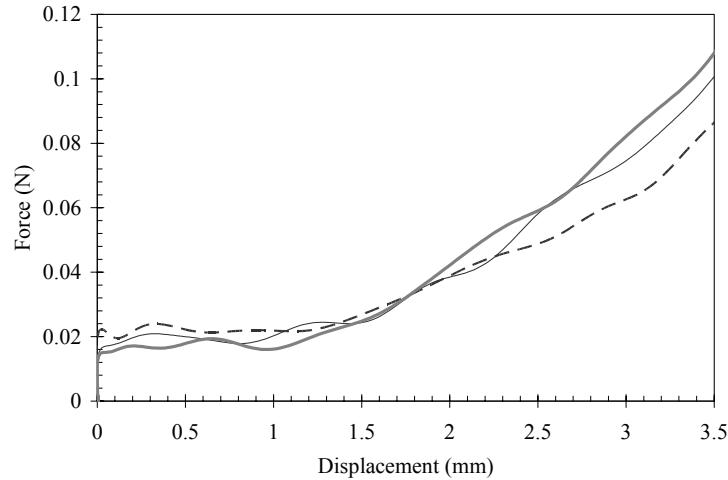


Figure 4: Force Response Variation by Location for Lung Deflection.

Figure 5 compares the kidney force response of a 48 day-old and 49 day-old porcine subject. Similar plots are shown for the liver in Figure 6 and for the lung in Figure 7. The mass of the liver in the 48 day-old swine was 37% greater than the liver of the 49 day-old swine. The kidney mass was 12% greater and the lung mass was 15% greater in the 48 day-old swine than in the 49 day-old swine. The kidney responses are similar with a mean force of 0.1658 N (3 mm of deflection) and a standard deviation of 0.0013 N (1% error). The mean force was 0.1247 N (3 mm of deflection) with a standard deviation of 0.0011 N (1% error) for the liver. The mean force was 0.0687 N (3 mm of deflection) with a standard deviation of 0.0092 N (13% error) for the lung.

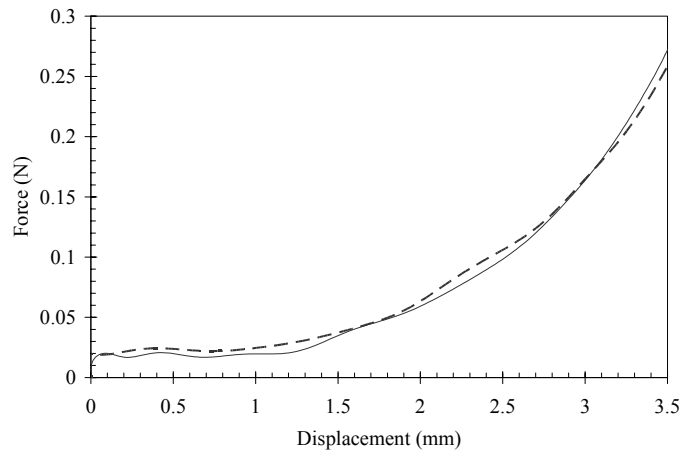


Figure 5: Force Response Variation across Subjects of Similar Age for Kidney Deflection.

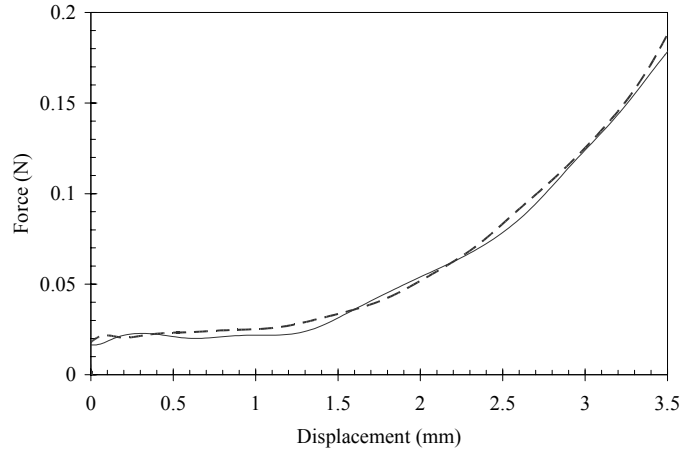


Figure 6: Force Response Variation across Subjects of Similar Age for Liver Deflection.

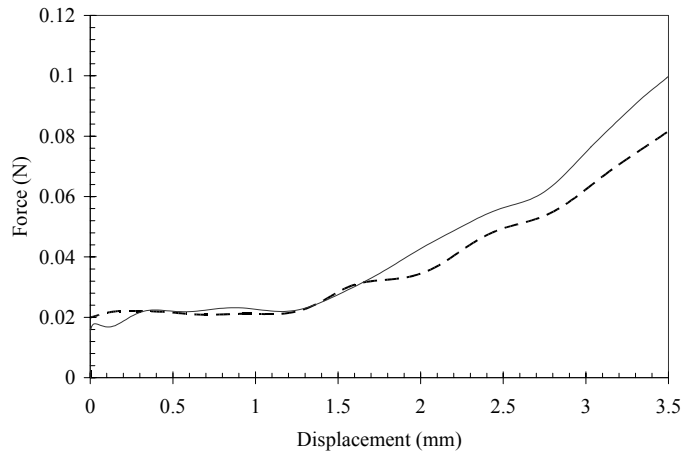


Figure 7: Force Response Variation across Subjects of Similar Age for Lung Deflection.

Figure 8 is a plot of the nonlinear force response of a 33 day-old and a 211 day-old kidney. The effective stiffness is measured at 3 mm of displacement. The K_{eff} is 0.0812 N for the 33 day-old and is 0.2506 N for the 211 day-old. Figure 9 plots the effective stiffness, K_{eff} , against the age of the subject for all kidney tests. Similar plots are shown for the liver (Figure 10) and for the lung (Figure 11). The equation for the linear model is given in each figure. It was determined that β was 0.0007 and α was 0.1068 for the kidney linear model (Figure 9), β was 0.0012 and α was 0.0716 for the liver (Figure 10), and β was -0.0002 and α was 0.0801 for the lung (Figure 11). The R^2 was 0.7049 and was significant at a p-value of less than 0.001 (Figure 9) for the linear model of the kidney. The R^2 was 0.6138 and was significant at a p-value of less than 0.004 for the liver (Figure 10). The R^2 was 0.3651 and was significant at a p-value of less than 0.022 for the lung (Figure 11).

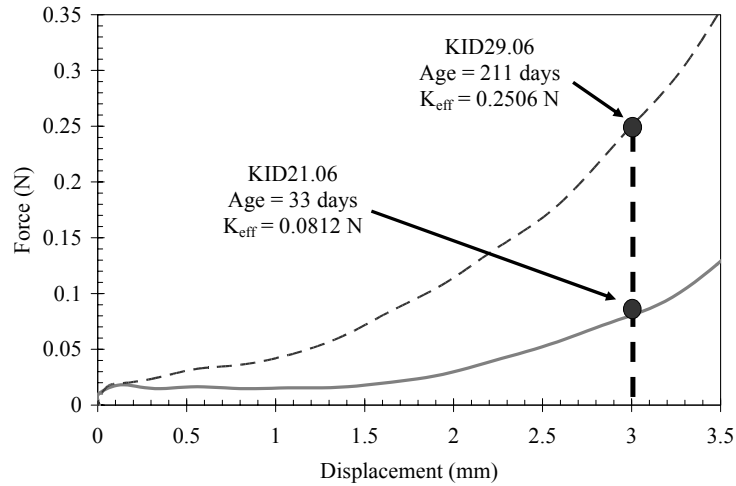


Figure 8: Variations in Kidney Force Response at 3 mm of Deflection for a Range of Test Subject Ages.

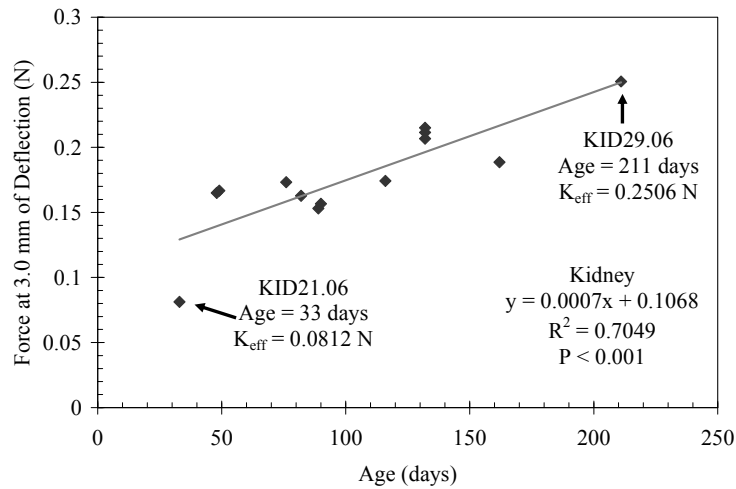


Figure 9: Variations in Kidney Force Response at 3 mm of Deflection for a Range of Test Subject Ages.

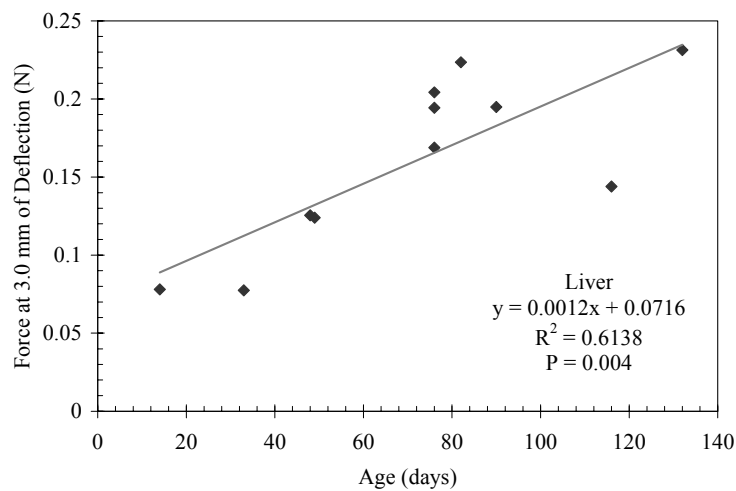


Figure 10: Variations in Liver Force Response at 3 mm of Deflection for a Range of Test Subject Ages.

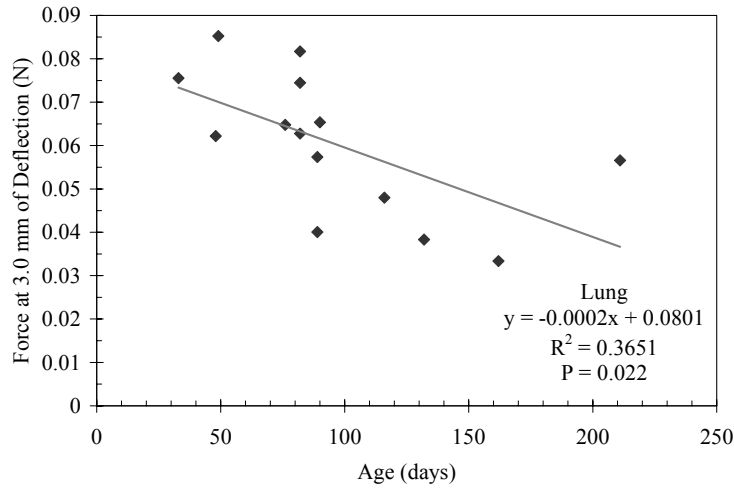


Figure 11: Variations in Lung Force Response at 3 mm of Deflection for a Range of Test Subject Ages.

DISCUSSION

The results presented above depend on the selection of a reasonable surrogate. To ensure that the age of our surrogates was representative of the age range of human pediatric development, an age correlation between the pig subjects and corresponding human subjects was made based on intact organ mass. Kayser (1987) listed the kidney, liver, and lung mass for humans ranging in age from infancy (1 day) to early physical adulthood (19 years). The porcine whole organ mass was compared to Kayser’s human organ mass and age relationship to generate a corresponding human age scale for our porcine specimens. The corresponding human age scale for the porcine organs ranged from 16 days to 19 years for the liver, 45 days to 19 years for the lung, and 150 days to 19 years for the kidney. The age range of the test subjects reasonably corresponds to the human age range representing pediatric development (infant to adult).

The findings presented in this paper evaluate the repeatability of the test setup and define trends for investigating changes in the force-penetration response for pediatric porcine organ development. The repeatability of the test fixture was validated by testing the same organ in different locations (Figures 2, 3, 4) and by comparing the responses of two organs of similar age (Figures 5, 6, 7). The force-penetration responses for three different kidney (Figure 2) locations are similar (2% error at 3 mm) and validate our test method, indicating that the organ location has a reasonably small effect relative to age. The plot comparing the liver force response for different organ locations (Figure 3) shows more variation (10% error at 3 mm) than the kidney, but some variation is expected due to inhomogenities such as bile and blood vessels throughout the liver. The fluctuations in the liver force response are assumed to be reasonable for this analysis and the test method is considered to be repeatable. The lung response showed the largest deviations compared to the magnitude of the mean response (13% error at 3 mm). This is assumed to be caused by the large changes in the size of the bronchial pathways throughout the lung.

Figure 5 compares the kidney force response of a 48 day-old and 49 day-old porcine subject. Despite the fact that the 48 day-old liver had a 12% greater mass than the 49 day-old liver, the responses are similar. This indicates that our test methodology is measuring the material response and not the structural response of the organ, which will allow us to compare results from organs of different sizes because the response is independent of organ mass. Figure 6 compares the liver force response for two subjects of similar ages. Once again, the plot shows that the responses are quite similar despite the 48% greater mass of the 48 day-old liver. Given the small differences in the response by location and across subjects, the assumption that the kidney and liver are a homogenous continuum seems reasonable. Figure 7 compares the force response for the lung across subjects of similar ages. The cross plot shows the largest deviations from

the mean response (13% error) which indicates the assumption that the lung is a homogeneous continuum begins to break down as the size of the bronchial tree becomes large relative to the parenchyma.

To investigate changes in the material response with age, the force response for each organ was measured at 3 mm of deflection yielding an effective stiffness for the organ. This penetration depth was chosen because it captures the nonlinearity (Figure 8) of the force response of each organ, yet seems to be small enough so that the effects of the substrate and organ thickness on the force response appear to be negligible. This analysis is merely an exploratory investigation of changes in the force response with age, and by no means to be considered a robust model of changes in material properties with age.

The linear models indicate that the effective stiffness for the kidney (Figure 9) and liver (Figure 10) increases with age, while the effective stiffness for the lung (Figure 11) decreases with age. Despite the problems with the homogeneous assumption for the lung and the deviations in the lung response, the negative correlation between lung effective stiffness at 3 mm of deflection and age was significant ($p = 0.022$) (Figure 11). The R^2 value for the kidney (Figure 9) linear model was 0.7049, indicating greater scatter in the response of the liver ($R^2 = 0.6138$) than in the kidney. The R^2 value of the lung (Figure 11) linear model was 0.3651, indicating that the measured effective stiffness for the lung has the largest scatter in the force response of the three organs. All three of these models are significant at a p-value of 0.05. These models apply only to porcine organs ranging in age from infancy to early adulthood. Given that there appear to be many similarities between the porcine and human anatomy, these results may indicate that the material properties of the pediatric human kidney, liver, and lung change with age.

The effective stiffness of the kidney and liver both increase with age. It is hypothesized that this may be due to an increase in the fibrotic tissue within the organs as the test subjects develop. A detailed histological analysis will be performed on samples taken from each test specimen to see if the presence of fibrotic tissue increases with age and to also investigate if any age-related microscopic structural changes exist. The effective stiffness of the lung decreases with age, which may be a structural response of the organ instead of a material response. Although the test setup appears to successfully evaluate changes in the material response of both the kidney and the liver, the structural components of the lung are quite different. As mentioned previously, the lack of homogeneity of the lung tissue may be responsible for the dissimilitude between subjects in the force response of the lung. It is not entirely clear why the force response of the lung differs from the kidney and liver, but material changes may be induced by respiration mechanics. This effect is hypothesized and should be investigated further in the future.

CONCLUSIONS

This research developed a method to quantify age-related changes in organ material response. The results of this study showed that the force response of the intact kidney, liver, and lung at 3 mm of deflection with a steel spherical punch changes with age. The age of the porcine subjects at the time the organs were tested ranged from 14 days to 211 days, which correlates to a human age range of approximately 16 days to 19 years based on subject anthropometry and organ mass relative to humans. The force response of the porcine kidney and liver showed a positive correlation with age, while the response of the lung showed a negative correlation with age. All trends were significant at a p-value of 0.05. The greatest variation in the force response was seen in the lung, then the liver, followed by the smallest variation in the kidney.

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APPENDIX

Test Number [‡]	Litter	(days) Age	(kg) Whole Body Mass	(g) Organ Mass	(cc) Organ Volume	(N) Force at 3 mm
KID-21.06	4	33	6.2	22.6	20	0.08123
KID-25.06	4	48	11.5	41.4	41	0.16494
KID-26.06	4	49	11.0	37.1	37	0.16674
KID-30.06	6	76	14.5	45.9	45	0.17316
KID-31.06	6	82	16.1	48.6	40	0.16274
KID-17.05	3	89	27.0	81.5	80	0.15306
KID-33.06	6	89	21.2	66.5	63	0.15662
KID-24.06	3	116	47.5	151.0	148	0.17420
KID-32.06	7	132	73.4	174.5	173	0.21502
KID-32.07A	7	132	73.4	174.5	173	0.21147
KID-32.07B	7	132	73.4	174.5	173	0.20660
KID-19.07	2	162	101.2	270.7	265	0.18858
KID-29.06	5	211	96.0	175.7	175	0.25061
LIV-18.05	4	14	3.8	130.0	110	0.07806
LIV-21.06	4	33	6.2	218.3	200	0.07751
LIV-25.03	4	48	11.5	484.0	440	0.12550
LIV-26.03	4	49	11.0	353.0	340	0.12390
LIV-30.03	6	76	14.5	495.0	460	0.20425
LIV-30.05	6	76	14.5	495.0	460	0.16889
LIV-30.06	6	76	14.5	495.0	460	0.19438
LIV-31.03	6	82	16.1	508.0	500	0.22345
LIV-33.03	6	89	21.2	700.0	650	0.19486
LIV-24.06	3	116	47.5	1490.0	1400	0.14401
LIV-32.03	7	132	73.4	1910.0	1850	0.23137
LUNG-21.04	4	33	6.2	39.2	75	0.07553
LUNG-25.03	4	48	11.5	68.9	100	0.06218
LUNG-26.03	4	49	11.0	60.1	85	0.07524
LUNG-30.03	6	76	14.5	87.0	100	0.06477
LUNG-31.03	6	82	16.1	82.6	90	0.07448
LUNG-31.05	6	82	16.1	82.6	90	0.06275
LUNG-31.06	6	82	16.1	82.6	90	0.08168
LUNG-17.02	3	89	27.0	249.8	250	0.05734
LUNG-17.06	3	89	27.0	249.8	250	0.04004
LUNG-33.03	6	89	21.2	109.6	110	0.06535
LUNG-24.04	3	116	47.5	290.1	475	0.04800
LUNG-32.03	7	132	73.4	362.1	563	0.03833
LUNG-19.05	2	162	101.2	315.9	550	0.03336
LUNG-29.03	5	211	96.0	389.5	500	0.05659

[‡] Test number format is as follows: Organ - Pig Number.Test Number

DISCUSSION

PAPER: **Age-Dependent Material Characterization of Porcine Abdominal Organs**

PRESENTER: **Jason Mattice, University of Virginia - Center for Applied Biomechanics**

QUESTION: *Guy Nusholtz, DaimlerChrysler*

Looking at your slides on histology [Yes], they look different to me when I look, looking at them, so I'm not exactly sure. Did you do some sort of comparison between a large number of livers and lungs or did you just take those two?

ANSWER: No. We're currently looking at histological slides from each specimen. The analysis is still being performed by Dr. Beatrice Lopez, but what we used was an initial analysis that—Let me get back to the slide that just shows. It shows that there's just similar structure for the human organs and the pig organs.

Q: Someone's taken the slide.

A: I know. Where does it go? There we go. Almost there. There we go. So, mainly what I wanted to show here is that were—there are differences and there are going to be between human and pig and actually, we've seen that there's more connective tissue in pig liver than in the human liver. But in general, we can see that the structures are similar, enough so that Dr. Beatrice Lopez thinks that the pig organs would be a good substitute for the human ones.

Q: Okay. So you didn't look at a whole range of different livers.

A: No. We're currently looking at that right now to see if there is a great variability in the differences with age.

Q: Okay. It looks that you haven't figured out exactly how to sort through the structural response from the material response. Are you considering developing a hierarchical model to address that or are you just going to use the lump mass models that you showed near the end?

A: Right now, we're in the preliminary stages of this research. I'd like to investigate that, but I haven't gotten to that point yet. So as of now, we're only looking at the lump mass model, but it would be interesting to see the development.

Q: And the question is: How do you mix the—You're going to have to have some sort of mesomodel of the connecting tissues and some sort of mesomodel, and you're going to have to have a method to mix them together so you can end up with a bulk model so you don't have a gazillion different elements.

A: Yeah. It's a complex task. But like I said right now, I'd like to get to that point, but we haven't gotten there yet. Right now, we're just investigating to see if there were, perhaps, in the response with age. And, I think what this shows is that it indicates that there may be. I mean, this is a focused analysis.

Q: But you don't know whether it's material or mesostructure. Okay.

A: No. I mean, we haven't isolated whether it's exactly material or it's structural.

Q: Okay. Thank you.

QUESTION: *Frank Pintar, Medical College of Wisconsin*

This is a great start on a tough subject. I'm a little—I'm interested in your, the way that you define growth and equate it to human equivalent. The animal development, as you know, the rate is much different. And so, did you just use mass itself of the organs?

ANSWER: Let me go to a slide here. We had a source that plotted the organ mass for ages of humans through pediatric development, actually. Here we go. Stockard & Dayner plotted these organ weights with age, and what we did is we measured that organ weight for each of our pig specimens and correlated that so we have two constraint functions to solve for the age-correlation between the two. So

we did it by organ mass for right now and we showed geometric similarity between our infant and our adult. And, we're assuming that that holds true, for this moment, just to say that the pig surrogate is a suitable substitute for the human.

Q: Now, did you account for the difference in rate of growth in terms of the pig versus the human, because the rates are probably quite different?

A: Yeah, the rates are different as we see a pig reach adult maturity within 150 days.

Q: So, did you do it as a percentage?

A: Yeah. We only compared it to mass. We had fit for human or child organ mass versus age and for pig organ mass versus age, and we compared that age. I mean, it's not linear as you can [Sure] see from the plot, but that's what we used to define our age range.

Q: Okay. Good.

QUESTION: *Erik Takhounts, NHTSA*

I have a similar question, semi-suggestion regarding your test methods. Probably suggestion is if you want to compare the ages, their material response between the two ages, that's fine. I think that's good because what you're doing: You're basically testing the local properties of the material, whatever that material is, I think it's your membrane that your testing, not that the material that's underlying the membrane too much. Because as you've shown in the kidneys, very homogenous, you are trying to prove us that it is actually homogeneous. And the fact that there is no difference shows you that your test methodology of testing homogeneity is not quite right. [Um hm.] When you talk about the absolute response—I'm going to use this test methodology to tell you what the material response of the kidney or the liver is, I don't think your methodology is applicable for the reason I just mentioned.

A: So the question is...?

Q: That's why I said it's semi-comment, semi-question.

A: Oh. Oh.

Q: You don't have to answer right away, just think about it.

A: No. I understand it. Thank you.

Q: Unless you have an answer.

A: At this time, I don't.

QUESTION: *John Melvin, Tandelta*

As Joe Bren said many years ago, working on the abdomen is sort of a can of worms, but it is a tough area. My question relates to what are your plans to deal with perfusion of these organs because it looks like your forces are pretty low here and they're fluid-filled organs, and the fluid filling has a big effect. We showed that back in the 70's when we did perfused organs and looking at the impact behavior, which is presumably what you're after although with belt loading it's not quite the same thing. There is big effect there.

ANSWER: No, I agree. There is. For this study, we're not trying to develop any sort of injury threshold. We're simply trying to look at material properties and we evaluated whether or not we should use organ perfusion, but we decided not to because we don't believe that the pressure added would be significant to change with age. We wanted to study actually the material itself in the absence of this pressurization to see if that changed with age. So what I'd like to do is: Now that we've seen this, seeing that there is some kind of a trend that might exist, is to go back and perform some kind of injury threshold testing using perfusion to get more accurate results that we could understand how the abdomen responds in a crash event.

Q: Yeah. I would think that the kidney in particular, with its very tough capsule, is basically—when it's fluid filled—you are going to have quite a structural effect, even under a low rate load.

A: I agree with you.

Q: Okay.