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Mechanical Characterization of Immature Porcine Brainstem in Tension at Dynamic Strain Rates

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Data Collection B
Statistical Analysis C
Data Interpretation D
Manuscript Preparation E
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Background: Many brain injury cases involve pediatric road traffic accidents, and among these, brainstem injury causes disastrous outcomes. A thorough understanding of the tensile characterization of immature brainstem tissue is crucial in modeling traumatic brain injury sustained by children, but limited experimental data in tension is available for the immature brain tissue at dynamic strain rates.

Material/Methods: We harvested brainstem tissue from immature pigs (about 4 weeks old, and at a developmental stage similar to that of human toddlers) as a byproduct from a local slaughter house and very carefully prepared the samples. Tensile tests were performed on specimens at dynamic strain rates of 2/s, 20/s, and 100/s using a biological material instrument. The constitutive models, Fung, Ogden, Gent, and exponential function, for immature brainstem tissue material property were developed for the recorded experimental data using OriginPro® 8.0 software. The *t* test was performed for infinitesimal shear modules.

Results: The curves of stress-versus-stretch ratio were convex in shape, and inflection points were found in all the test groups at the strain of about 2.5%. The average Lagrange stress of the immature brainstem specimen at the 30% strain at the strain rates of 2, 20, and 100/s was 273±114, 515±107, and 1121±197 Pa, respectively. The adjusted R-Square (R^2) of Fung, Ogden, Gent, and exponential model was $0.820 \le R^2 \le 0.933$, $0.774 \le R^2 \le 0.940$, $0.650 \le R^2 \le 0.922$, and $0.852 \le R^2 \le 0.981$, respectively. The infinitesimal shear modulus of the strain energy functions showed a significant association with the strain rate ($p<0.01$).

Conclusions: The immature brainstem is a rate-dependent material in dynamic tensile tests, and the tissue becomes stiffer with increased strain rate. The reported results may be useful in the study of brain injuries in children who sustain injuries in road traffic accidents. Further research in more detail should be performed in the future.

MeSH Keywords: Brain Injuries • Brain Stem • Viscoelastic Substances

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Background

With a rapid rise in motor vehicle use around the world, large numbers of injuries and deaths due to road traffic accidents (RTAs) occur annually, as detailed in a report of World Health Organization (WHO) [1]. In some countries children account for a large proportion of traffic injuries. For example, although children aged 15 years and younger account for about 18% of the population in some developed countries, children injured in crashes account for about one-third of the pedestrian accident dataset [2].

Brain injury is still a leading cause of death and disability for children and adolescents involved in RTAs. A report by Langlois et al. [3] showed that approximately 475 000 TBIs occurred each year among children aged 0 to 1 years. It was suggested that trauma to the brainstem is a hallmark of severe head injury because of its important role in physiological function. The mechanism of brain injury has been discovered to substantially involve impact to or rotation of the head, causing deformation of the brain, in which brain injury is associated with strain and strain rate. Owing to the geometry of the central nervous system (CNS), the brainstem is vulnerable to the high strain induced by the combination of compression, tension, and shear, especially in rotational acceleration loading on the head [4].

Head computational models are powerful tools for use in studying adult traumatic brain injury (TBI) mechanisms and determining adult brain injury tolerance [5]. To discover age-pertinent mechanisms and estimate the injury threshold of children and young adults, models are needed for use in studying pediatric brain injuries [6]. Such tools depend heavily on knowledge of pediatric brain tissue material properties and anatomical structures. Owing to the paucity in basic data on the material properties of pediatric brain tissue and skull, the child head finite element model was developed, mostly by scaling the adult head models, in which the child model was regarded as a miniature adult model. However, recent studies show age-related differences in the brain [7,8], meaning that it is not reasonable to regard the child's head as a miniature adult head.

There have been many studies concerning brain tissue properties. For example, compressive and tensile tests of brain tissue at quasi-static strain rates were done to study the tissue mechanical properties under surgical conditions owing to considerably lower strain rate [9,10]. Other studies were performed to address the mechanical properties of brain tissue during impact events with compression tests at strain rates of 1, 10, and 50/s and tension tests at strain rates of 0.9, 4.3, and 25/s [11,12]. In the experiments involving brain tissue compression and tension by Rashid et al. [13,14], the 30% strain rates were 30, 60, and 90/s. Numerous studies have reported the adult brain tissue mechanical properties associated with brain injury in experiments at over 20% strain at strain rats of between 1 and 100/s [13–18]. A few studies have reported on the material properties of immature brain tissue: Prange [19], Prange and Margulies [8], Duhaime et al. [20], Ning et al. [21], and Li et al. [22]. Consequently, the brain material properties of children are not well understood, especially the tensile behavior.

Due to ethics constraints, there is great difficulty in obtaining fresh brain tissue from children; therefore, in the present study we used immature porcine brain tissue to serve as a substitute to study pediatric brain tensile behavior. The aim of this study was to conduct tensile tests in immature brainstem specimens at high stretch velocity (i.e., dynamic strain rates) and then determine the mechanical behavior of the immature brainstem during traumatic events.

Material and Methods

The Research Ethics Committee of the Third Military Medical University granted ethics approval prior to the tests. All the tests were performed strictly following the rules listed in the Guide for the Care and Use of Laboratory Animals, as confirmed by the U.S. National Institutes of Health. Great care was taken to reduce suffering of the animals.

Specimen preparation

A total of 23 immature pigs from the same farm, aged 4 weeks (at a developmental stage similar to human toddlers), with an average weight of 16±0.5 kg, were killed at a local slaughter house (Chongqing BORN Biological Technology Co., Ltd.). The brain was harvested immediately from the pigs as byproduct, and then stored in a thermally insulated stainless steel vessel filled with a saline solution with 0.9% NaCl at 4–50°C to maintain ionic balance during transportation to our lab.

The cerebrum, cerebellum, and brainstem were separated in the lab, and then the midbrain of the brainstem was cut into a plate with a thickness of 5 mm along the axis of the upper brainstem. A steel circular trephine of a diameter of 9 mm with sharp edges was used to cut the brainstem plate to produce a specimen with a length of 5 mm and a diameter of 9 mm. In this section the main fibers of the brainstem were considered to orient along the long axis of the cylinder, as illustrated in Figure 1. During the preparation, the samples were sprayed with the physiological solution to maintain the *in vitro* physical environment.

Experimental study

To perform the tensile experiment, we used a computer-controlled, high test precision Bose ElectroForce 3100 machine

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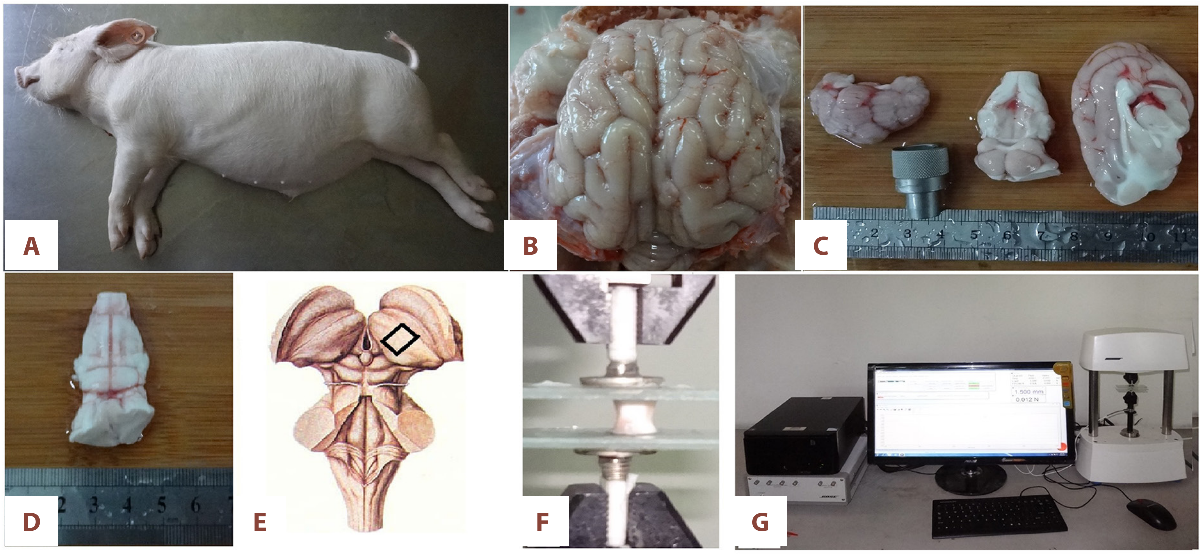

Figure 1. Photos of specimen preparation, dynamic tensile machine. (**A**) An immature pig. (**B**) Fresh porcine brain harvested as a byproduct. (**C**) Separated infant porcine cerebrum, cerebellum, brainstem, and the steel circular cylindrical trephine used to make specimens. (**D**) Infant porcine brainstem tissue. (**E**) Schematic diagram for the anatomical location of the cylindrical brainstem sample, indicated in the black rectangle area. (**F**) Porcine brainstem tissue sample experienced extension. (**G**) The biomaterial test machine.

(Bose Corporation, Gillingham, UK), adapted for testing biological specimens. The instrument contains an electromagnetic motor with a stroke resolution of 0.0015 mm and a maximum stroke length of 5 mm, and a minimum load resolution of 6 mN with a 22N load cell. The equipment was designed to have a high response speed so that the configured loading speed can be achieved instantly. The history of stretch and force experienced by the tissue were recorded when performing tensile tests.

Experimental protocol

Owing to the great challenge in gripping the very soft biological tissue in the tensile test [10,23], great care was taken during preparation and testing of the specimens. We used tensile grips and clips of the apparatus to rigidly fix 2 glass slides in which the one fixed to the stroke was considered as the top, and the one fixed to the load cell was regarded as the bottom. A thin layer of surgical glue (Cyanoacrylate, low-viscosity Z105880-1EA, Sigma-Aldrich) was used to firmly cover the surface of both slides. The prepared tissue specimen was carefully placed on the bottom slide, and then the stroke was slowly lowered to slightly touch the superior surface of the specimen. The stroke was stopped when the load reached 5 mN. To ensure proper adhesion of the specimen to the top and bottom platen, the stroke would stay for 1 min prior to the tensile test.

After super- and sub-surfaces of the tissue were fixed to both slides using the procedure detailed above, the tests were carried out at stroke velocities of 10, 100, and 500 mm/s, in which the strain rate of the specimen was 2, 20, and 100/s, respectively. For all the test specimens, no preconditioning was done, and each sample only experienced 1 loading cycle. The final status of the specimens was checked when finishing the test, and the data were not used in subsequent analysis if the glue between the slides and faces of the specimen failed. All the tests were finished at room temperature of 22°C within 6 h postmortem.

Data processing and analysis

The nominal stress, S_{11}, was calculated using Formula (1), in which F represents the tension force in newtons, and A was the cross-section area of tested tissue in m^2.

$$S_{11} \equiv \frac{F}{A} \tag{1}$$

From the conclusion drawn by Miller [23], the stretch ratio λ for the specimen in the tensile test with confined conditions is related to the height change of the tissue if a value of $\frac{h}{H}$ is within 1.0~1.3, so

$$\lambda = K\left(\frac{h}{H} - 1\right), K = 1.583 \tag{2}$$

The constitutive models of immature brainstem tissue may be developed based on quasilinear viscoelastic (QLV) theory [24,25], in which the relationship between stress and strain tensors was derived from a strain energy potential function.

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The function, W, is defined using the invariants (I_1, I_2, I_3) of the strain tensor. The third strain (I_3) is unity, as the tissue is incompressible. As a result, the function is described in simplified form using I_1 and I_2. The invariants, I_1 and I_2, are defined as:

$$I_1 = \lambda_1^2 + \lambda_2^2 + \lambda_3^2 \quad (3)$$

$$I_2 = \lambda_1^2\lambda_2^2 + \lambda_2^2\lambda_3^2 + \lambda_3^2\lambda_1^2 \quad (4)$$

In the formulas, the squares of the principal stretch ratios, λ_2^1, λ_3^2, and λ_3^2, had the relationship of $\lambda_2^1\lambda_3^2\lambda_3^2$=1, due to incompressibility [26]. As assumed homogeneous deformation conditions of the brain tissue under tension, the Eulerian and Lagrangian principal axes of strain and stress are aligned with the tensile directions, with any 2 orthogonal axes x_2, x_3. Hence, the stretch ratios are indicated by the formula:

$$\lambda_1 = \lambda \text{ and } \lambda_2 = \lambda_3 = \frac{1}{\sqrt{\lambda}} \quad (5)$$

In the extensional test, the value of λ is not less than 1, so I_1 and I_2 are expressed by the formula:

$$I_1 = \lambda^2 + 2\lambda^{-1} \text{ and } I_2 = \lambda^{-2} + 2\lambda \quad (6)$$

Then, the stresses derived from the experiments may be compared to the predicted using the hyperelastic models from the formula:

$$S_{11} = \frac{dw}{d\lambda}, \text{ where } W(\lambda) \equiv W(\lambda^2 + 2\lambda^{-1}, \lambda^{-2} + 2\lambda) \quad (7)$$

The material parameters of some common hyperelastic models (e.g., Fung, Gent, and Ogden) may be obtained from fitting the curves.

Widely used to model soft biological tissues in tension, the Fung isotropic energy function depends on the first strain invariant only:

$$W = \frac{\mu}{2b}\left[e^{b(I_1-3)} - 1\right] \quad (8)$$

yielding the following nominal stress component S_{11} along the x_1-axis:

$$S_{11} = \mu e^{b(\lambda^2+2\lambda^{-1}-3)}(\lambda - \lambda^{-2}) \quad (9)$$

where infinitesimal shear modulus μ(>0) and stiffening parameter b(>0) are the 2o constant material parameters [27].

Gent strain energy function, widely used to describe viscoelastic material, also depends on the first strain invariant only:

$$W = -\frac{\mu}{2}J_m \ln\left(1 - \frac{I_1-3}{J_m}\right) \quad (10)$$

then,

$$S_{11} = \frac{\mu J_m}{J_m - \lambda - 2\lambda^{-2} + 3}(\lambda - \lambda^{-2}) \quad (11)$$

in which infinitesimal shear modulus μ>0 and J_m>0) are the 2 constant material parameters [28].

Ogden strain energy function is given by:

$$W = -\frac{2\mu}{\alpha^2}(\lambda_1^\alpha + \lambda_2^\alpha + \lambda_3^\alpha - 3) \quad (12)$$

producing the stress form:

$$S_{11} = \frac{2\mu}{\alpha}\left\{\lambda^{\alpha-1} - \lambda^{-(\frac{\alpha}{2}+1)}\right\} \quad (13)$$

in which μ>0 is the infinitesimal shear modulus, and α is a stiffening parameter [29].

In addition, an exponential model was used to fit the Lagrange stress-versus-stretch ratio shown by the following formula:

$$S_{11} = S_0 + Ae^{-\lambda/t0} \quad (14)$$

All operations for fitting were done using the software OriginPro® 8.0. The t test was performed for infinitesimal shear modulus, μ, in which a value of $p<0.05$ was considered to be significant.

Results

The history curve of Lagrange stresses for the tested brainstem specimens determined according to the formula (1) are illustrated in Figure 2. The brainstem tissue stretch ratio λ was determined using equation (2) from the distance that the tissue was stretched. The curves of the stress-versus-stretch ratio shown in Figure 3 were convex in shape. The average Lagrange stress for the tested specimen at the 30% strain at the strain rates of 2, 20, and 100/s were 273.3±114, 515±107, and 1121±197 Pa, respectively.

From the curves of Lagrange stress-versus-stretch ratio illustrated in Figure 3, it was found that the material became stiffer with an increase in strain rate. An interesting phenomenon was that all curves indicated points of inflection at the tensile strain of about 2.5% in each test group.

The Lagrange stress-against-stretch ratio, λ, can be fitted with the strain energy function of Fung ($0.820 \le R^2 \le 0.933$), Ogden ($0.774 \le R^2 \le 0.940$), and Gent ($0.650 \le R^2 \le 0.922$), and the exponential model ($0.852 \le R^2 \le 0.981$), as shown in formulas (15–17). Figure 4 exhibits curve fitting of Fung, Ogden, and Gent strain energy function, and the exponential model for the average

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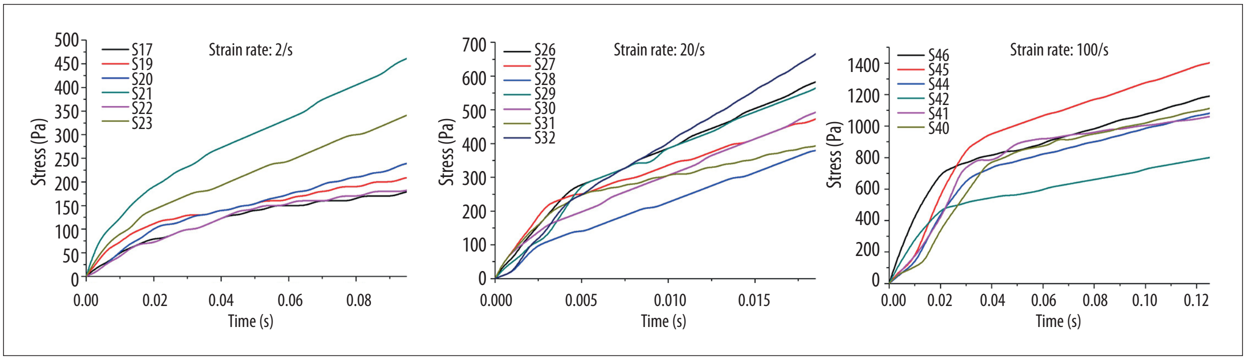

Figure 2. The history curve of Lagrange stress for the infant porcine brainstem specimen at the dynamic tensile tests. A total of 6 tests were conducted successfully at the stretch speed of 10 mm/s (strain rate 2/s), 7 at the stretch speed of 100 mm/s (strain rate 20/s), and 6 at the stretch speed of 500 mm/s (strain rate 100/s).

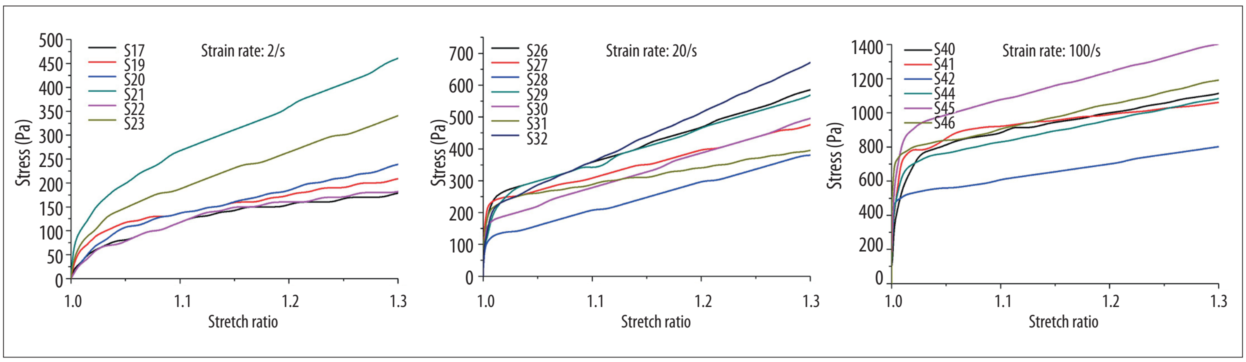

Figure 3. The curve of Lagrange stress-versus-stretch ratio for the infant porcine brainstem specimen at the dynamic tensile tests. A total of 6 tests were conducted successfully at the stretch speed of 10 mm/s (strain rate 2/s), 7 at the stretch speed of 100 mm/s (strain rate 20/s), and 6 at the stretch speed of 500 mm/s (strain rate 100/s).

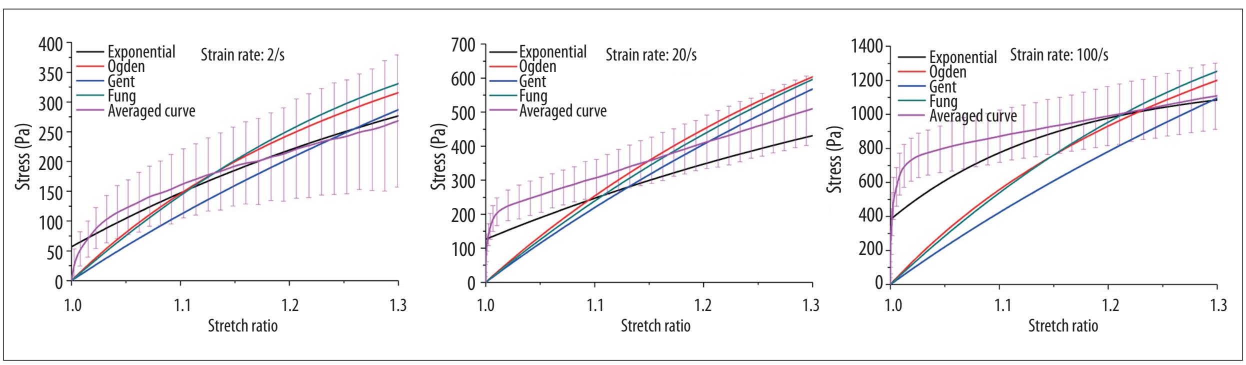

Figure 4. The curve fitting of Fung, Ogden, and Gent strain energy function, and exponential model for the average experimentally acquired data of stress-versus-stretch ratio at each test group. The adjusted R-Square of the model fitting the experimental data up to the strain of 30% at the dynamic strain rate is $0.820 \leq R^2 \leq 0.933$ in Fung, $0.774 \leq R^2 \leq 0.940$ in Ogden, $0.650 \leq R^2 \leq 0.922$ in Gent, and $0.852 \leq R^2 \leq 0.981$ in exponential model.

curve experimentally acquired. We noted that the change trend of stress increase at the 2.5% tensile strain did not agree well with the fitted models:

$$S_{11}=499.9-4348.4e^{-\lambda/0.438} \tag{15}$$

$$S_{11}=845.8-10187.8e^{-\lambda/0.377} \tag{16}$$

$$S_{11}=1200-8377460.0e^{-\lambda/0.108} \tag{17}$$

Table 1 lists the infinitessimal shear modulus, μ, derived from the fitted strain energy function, Fung, Ogden and Gent, in which the infinitessimal shear modulus, μ, was found to have a strong rate dependence. For Ogden model in the table, for instance, the infinitessimal shear modulus at the strain rate

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Table 1. The infinitessimal shear modulus, μ, of Fung, Ogden, and Gent function at dynamic strain rates (mean ±SD and μ>0).

Strain rate/s	Strain energy function (Unit: Pa)		
	Fung	Ogden	Gent
2	543.0±252.1*	545.2±210.3*	691.1±202.8
20	882.9±135.2*,**	964.2±178.3*,**	802.2±163.9*
100	2010.7±315.3**	2281.1±503.2**	1547.0±348.7*

*,**: $p<0.01$.

of 100/s seemed to be 4.2 times higher than that at the strain rate of 2/s.

Discussion

Road traffic injuries will increase to become the fifth leading cause of death by 2030 and child safety recently has become an important research field around the world according to the WHO [1,2]. Head injuries are common in children involved in RTAs [2,3]. To protect children from injury and death, therefore, it is necessary to thoroughly understand the mechanisms of TBI prior to developing the measurements to reduce and eliminate the injury.

The finite element model, as a powerful tool, may play a significant role in discovering injury mechanisms in the human body, in which injury response and tolerance could be determined [5,6,30]. The characteristics of brain material mechanical properties are of importance in developing accurate computational models, contributing to an increasing research focus on brain tissue mechanical characterization [6,30,31], and many adult brain tissue property studies have been carried out [7,8,11,15]. However, few studies have been performed to investigate pediatric brain tissue properties, especially for the tensile test, because of challenges in the experimental techniques [31]. It should be noted that mechanical characterization of brain tissue at high loading velocities is crucial for modeling TBI. Brain tissue properties in tension are not as well defined as in other loading modes, such as compression and shear. Therefore, we carried out a series of tests to study the tensile material property of the brainstem.

Methodological issues could contribute to the apparently disparate material properties reported in some published studies [31] and the experimental technology was regarded as the greatest challenge for tensile tests in very soft biological tissue [24]. Much of the large disparity in previously reported data may be explained by the more rigorous approaches to control of sample preparation, test conditions, and the test procedures [31]. In the present study, the brain samples were uniform and came from the same source, the experimental protocol remained consistent, and great care was taken when performing the tensile tests, while there was a huge disparity in each test group, as shown in Figures 2 and 3.

Because of the paucity of data on pediatric brainstem tissue, the present results are difficult to directly compare with previous reports. The present results show that the average Lagrange stress for the tested brainstem specimen at the 30% tensile strain at the strain rate of 2, 20, and 100/s was 273±114, 515±107, and 1121±197 Pa, respectively. In the results of Miller and Chinzei [10], at the strain rate of 0.64/s and 0.0064/s, the stress sustained by the tested brain specimen was lower than 300 Pa, while Rashid et al. [14] reported that under tensile tests at the dynamic strain rates of 30/s, 60/s, and 90/s, the stresses were over 3000 Pa.

Similar to the material behavior of adult brain specimens found in some previous reports [13,32–36], the immature brainstem tissue showed a significant rate dependence in dynamic tensile tests in the present study. With an increase of the strain rate in the tensile tests, the immature porcine brainstem tissue became stiffer. Using porcine brain tissue, Thibault and Margulies [37] performed the first study of the immature brain, reporting a significant increase in the complex shear modulus of cortical gray matter obtained from 2–3-day-old piglets.

Some published data on brain tissue material properties at low strain rate show the brain tissue becomes softer with strain increase [10,38], but another study reported that at dynamic strain rate the brain tissue became stiffer with an increase of the strain [13]. In the present study, the immature porcine brainstem tissue became softer with strain increase, in which all the curves of stress-versus-stretch ratio were convex in shape. The exponential model seems to best match the immature brainstem tissue material property data in the tensile tests compared to the strain energy function. In addition, we noted that there were significant inflection points at the strain of 2.5% for all the test groups. We presumed that the linear viscoelastic regime of immature porcine brainstem in dynamic tensile tests was within the tensile strain of 2.5%. However, all the models developed for the immature porcine brainstem tissue did not fit the multi-stage experimental data combining the linear regime and non-linear regime, and another model needs to be developed to fit the immature brain material property of multi-stage characterization.

Our results show that the immature porcine brainstem had strong rate dependence, while the estimated shear modulus increased with an increase of the strain rate. In addition, we found that the shear modulus of immature brainstem tissue in the present study was higher than that of immature cerebellum tissue previously determined in the lab following the same procedures [22]. By conducting oscillatory shear tests, Arbogast and Margulies [5] reported that the complex modulus and its 2 components, the storage and loss modulus, varied with testing frequency, indicating a viscoelastic response. In their results, the experimentally determined shear modulus of the brainstem was 2 orders of magnitude lower than 168 kPa. Compared with the brain test data in the same lab [39], the brainstem has more complex modulus, particularly for the storage modulus component [5].

Computer models of the pediatric head commonly use ratios of adult-to-juvenile porcine brain material properties to extrapolate pediatric material properties from human adult material properties because of the paucity of data on the properties pediatric human brain tissue [6,30]. It should be noted that age-dependent material properties of brain tissue are not linear, but rather rapidly change during the first few years of life, and then change more gradually in early childhood. Axons, rather than the surrounding matrix of astrocytes and oligodendrocytes, contribute more to the stiffness of brain tissue according to some previously reported biomechanical models [21,31]. Axons in the pediatric brain undergo rapid myelination during the first year of life and the pediatric brain reaches adult levels of myelination at approximately 18 months old; therefore, the composition of the 5-year-old brain is more similar to that of an adult brain. In the present study, only 4-week-old pigs were chosen to test the brainstem material properties, in which the material property of the immature brainstem tissue was carried out in 1 direction, and only the tensile experiment, without the shear and compressive tests, was performed. Further research is needed to improve our understanding of the details of brainstem mechanical behavior.

Limitations

We attempted to describe the material properties of immature porcine brainstem tissue at dynamic strain rates to build a finite element model of the pediatric brain. However, our study has some limitations that need to be mentioned. First, the number of samples and valid data for studying the material properties of brainstem tissue from immature pigs was limited. Furthermore, pigs aged 4 weeks are similar to human infants, which only account for a small proportion of all children. In high-rate tests, there is a period in which the crosshead is ramping up to the specified speed, and it is not exactly clear how long it took to achieve this speed in our study. The brainstem is anisotropic and therefore may violate the assumption of Miller [23] that the materials are isotropic and incompressible. In this study, only tensile tests were done, and it would be better if the compression and shear tests could also be performed in discover the material properties of the brain. Finally, immature porcine brain tissue served as a substitute to study the pediatric material tensile behavior in the current study, but a significant difference may exist in material properties between pigs and humans.

Conclusions

To address the material properties of immature brainstem in tension, dynamic tensile tests at 3 stretch speeds were performed for the brainstem tissue specimens obtained from 4-week-old pigs in the present study. Although some limitations exist in our study, the results show that the immature brainstem is a rate-dependent material in dynamic tensile tests, and the tissue becomes stiffer with an increase of strain rate, with inflection points in all the test groups at the strain of about 2.5%. The function of Fung, Ogden, Gent, and exponential model may match well with the material behavior of the brainstem for immature pigs in dynamic tensile testing. The reported results may be useful in understanding brain injuries in children who have been in road traffic accidents.

References:

1. World Health Organization: Global status report on road safety 2013: Supporting a Decade of Action. World Health Organization, 2013
2. Mizuno Y: Summary of IHRA pedestrian safety WG activities (2005) – proposed test methods to evaluate pedestrian protection afforded by passenger cars. 19th ESV. 2005; 05-0138
3. Langlois J A, Rutland-Brown W, Thomas KE: The incidence of traumatic brain injury among children in the United States. J Head Trauma Rehabil, 2005, 20(3): 229–38
4. Kristy B Arbogast, Margulies SS: Material characterization of the brainstem from oscillatory shear tests. J Biomech, 1998, 31(9): 801–7
5. Mao H, Yang KH: Investigation of brain contusion mechanism and threshold by combining finite element analysis with *in vivo* histology data. Int J Numer Method Biomed Eng, 2011; 27(3): 357–66
6. Crandall JR, Myers BS, Meaney DF et al: Pediatric injury biomechanics. Springer Berlin, 2012; 59: 44–49
7. Brittany C, Margulies SS: Material properties of human infant skull and suture at high rates. J Neurotrauma, 2006; 23(8): 1222–32
8. Prange MT, Margulies SS: Regional, directional, and age-dependent properties of the brain undergoing large deformation. J Biomech Eng, 2002; 124(2): 244–52

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9. Miller K, Chinzei K: Constitutive modelling of brain tissue: Experiment and theory. J Biomech, 1997; 30(11–12): 1115–21
10. Karol M, Kiyoyuki C: Mechanical properties of brain tissue in tension. J Biomech, 2002; 35(4): 483–90
11. Tamura A, Hayashi S, Nagayama K et al: Mechanical characterization of brain tissue in high-rate extension. Journal of Biomechanical Science and Engineering, 2008; 3(2): 263–74
12. Tamura A, Hayashi S, Watanabe I: [635 Mechanical characterization of porcine brain tissue in high-rate compression.] 2007; 2006(3): 452–53 [in Japanese]
13. Rashid B, Destrade M, Gilchrist MD: Mechanical characterization of brain tissue in compression at dynamic strain rates. J Mech Behav Biomed Mater, 2012; 10: 23–38
14. Rashid B, Destrade M, Gilchrist M D: Mechanical characterization of brain tissue in tension at dynamic strain rates. J Mech Behav Biomed Mater, 2014, 33(5): 43–54.
15. Bain AC, Meaney DF: Tissue-level thresholds for axonal damage in an experimental model of central nervous system white matter injury. J Biomech Eng, 2001; 122(6): 615–22
16. Geddes DM, Cargill RS: An *in vitro* model of neural trauma: device characterization and calcium response to mechanical stretch. J Biomech Eng, 2001; 123(3): 247–55
17. Pfister BJ, Weihs TP, Betenbaugh M et al: An *in vitro* uniaxial stretch model for axonal injury. Ann Biomed Eng, 2003; 31(5): 589–98
18. Bayly PV, Black EE, Pedersen RC et al: *In vivo* imaging of rapid deformation and strain in an animal model of traumatic brain injury. J Biomech, 2006; 39(6): 1086–95
19. Prange M: Biomechanics of traumatic brain injury in the infant. Ph.D. thesis, Department of Bioengineering. University of Pennsylvania, PA. 2002; 186
20. Duhaime AC, Margulies SS, Durham SR et al: Maturation-dependent response of the piglet brain to scaled cortical impact. J Neurosurg, 2009; 93(3): 455–62
21. Ning X, Zhu Q, Lanir Y et al: A transversely isotropic viscoelastic constitutive equation for brainstem undergoing finite deformation. J Biomech Eng, 2006; 128(6): 925–33
22. Kui L, Hui Z, Wenjun L et al: Material properties and constitutive modeling of infant porcine cerebellum tissue in tension at high strain rate. PLoS One, 2015; 10(4): e0123506
23. Miller K: How to test very soft biological tissues in extension? J Biomech, 2001; 34(5): 651–57
24. Fung YC: Mechanical properties of living tissues. 2nd ed. Journal of Applied Mechanics, 1993; 49(2): 1306–9
25. Ogden RW: Non-linear elastic deformations. Courier Corporation, 1997
26. Fung Y C, Fronek K, Patitucci P: Pseudoelasticity of arteries and the choice of its mathematical expression. Am J Physiol, 1979; 237(5): H620–31
27. Fung YC: Elasticity of soft tissues in simple elongation. Am J Physiol, 1967; 213(6): 1532–44
28. Gent AN: A new constitutive relation for rubber. Rubber Chemistry & Technology, 1996, 69(1): 59–61
29. Ogden RW: Large deformation isotropic elasticity-on the correlation of theory and experiment for incompressible rubberlike solids. Proceedings of the Royal Society of London, 1972; 326(1567): 565–84
30. Coats B, Margulies SS, Ji S: Parametric study of head impact in the infant. Stapp Car Crash Journal, 2007, 51
31. Miller K: Biomechanics of the brain. Biological & Medical Physics Biomedical Engineering, 2011; 12(2): 189–219
32. Kristy B Arbogast, Margulies SS: Fiber-reinforced composite model of the viscoelastic behavior of the brainstem in shear. J Biomech, 1999; 32(8): 865–70
33. Bilston LE, Liu Z, Phan-Thien N: Large strain behaviour of brain tissue in shear: some experimental data and differential constitutive model. Biorheology, 2001; 38(4): 335–45
34. Lippert SA, Rang EM, Grimm MJ: The high frequency properties of brain tissue. Biorheology, 2004; 41(6): 681–91
35. Jin X, Lee JB, Leung LY et al: Biomechanical response of the bovine pia-arachnoid complex to tensile loading at varying strain-rates. Stapp Car Crash Journal, 2006; 50: 637–49
36. Begonia MT, Prabhu R, Liao J et al: The influence of strain rate dependency on the structure-property relations of porcine brain. Ann Biomed Eng, 2010; 38(10): 3043–57
37. Thibault KL, Margulies SS: Age-dependent material properties of the porcine cerebrum: Effect on pediatric inertial head injury criteria. J Biomech, 1999; 31(12): 1119–26
38. Velardi F, Fraternali F, Angelillo M: Anisotropic constitutive equations and experimental tensile behavior of brain tissue. Biomech Model Mechanobiol, 2006; 5(1): 53–61
39. Thibault KL: Pediatric Head Injuries: The influence of brain and skull mechanical properties. Ph.D. Thesis, University of Pennsylvania, 1997

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