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**MODELING OF THE NON-AUDITORY
RESPONSE TO BLAST OVERPRESSURE**

**Gastrointestinal Tract Blast Injury
Laboratory Test Techniques**

ANNUAL/FINAL REPORT

**James H.-Y. Yu
Edward J. Vassel
James H. Stuhmiller**

JANUARY 1990

Supported by

**U.S. ARMY MEDICAL RESEARCH AND DEVELOPMENT COMMAND
Fort Detrick, Frederick, Maryland 21701-5012**

Contract No. DAMD17-85-C-5238

**JAYCOR
11011 Torreyana Road
San Diego, California 92121-1190**

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19. ABSTRACT (*Continued from front*)

GASTROINTESTINAL TRACT BLAST INJURY LABORATORY TEST TECHNIQUES

James H.-Y. Yu
Edward J. Vasel
James H. Stuhmiller
Applied Science and Engineering Technology
JAYCOR

ABSTRACT

Gastrointestinal tract blast injury involves the interaction of the physical aspect of the blast loading, the dynamics of bubble deformation/oscillation, and the physiological properties of the G.I. tract injury threshold. Such interplay is complicated by the random distribution of bubble size and bubble location. Because of these complications, there are several technical hurdles to be overcome before standardized test procedures can be established. JAYCOR has developed several techniques that facilitate the experimental effort. These include: (1) the design of a constant temperature test chamber system that can be used to deliver an equivalent intra-abdominal blast signal and facilitate high speed photographic observation of local G.I. deformation; (2) an autologous in vitro perfusion technique that allows a complete viable gastrointestinal tract to be isolated in a test chamber for blast injury tests, while under the continued blood supply from the cardiopulmonary system of the same test animal; (3) a probe-in-balloon unit that permits gas bubble volume to be placed at desired locations in a G.I. tract for measurement of its corresponding pressure signals; (4) an ultrasonic sensing technique for determining the location and length of gas bubbles in the G.I. tract; and finally, (5) techniques for indicating the presence, and quantifying the degree, of serosal and mucosal bleeding by Hemastix and microscopic examination.



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ABSTRACT

Gastrointestinal tract blast injury involves the interaction of the physical aspect of the blast loading, the dynamics of bubble deformation/oscillation, and the physiological properties of the G.I. tract injury threshold. Such interplay is complicated by the random distribution of bubble size and bubble location. Because of these complications, there are several technical hurdles to be overcome before standardized test procedures can be established. JAYCOR has been under contract with Walter Reed Army Institute of Research to perform small animal blast injury tests to support computational effort for blast injury prediction, and to assist field test instrumentation and data analysis. Through these efforts several techniques that facilitated the experimental effort were developed. These include: (1) the design of a constant temperature test chamber system that can be used to deliver an equivalent intra-abdominal blast signal and facilitate high speed photographic observation of local G.I. deformation; (2) an autologous in vitro perfusion technique that allows a complete viable gastrointestinal tract to be isolated in a test chamber for blast injury tests, while under the continued blood supply from the cardiopulmonary system of the same test animal; (3) a probe-in-balloon unit that permits gas bubble volume to be placed at desired locations in a G.I. tract for measurement of its corresponding pressure signals; (4) an ultrasonic sensing technique for determining the location and length of gas bubbles in the G.I. tract; and finally, (5) techniques for indicating the presence, and quantifying the degree, of serosal and mucosal bleeding by Hemastix and microscopic examination.

1. INTRODUCTION

It is known that strong or repeated blast overpressures can cause internal organ injuries [1]. Slight to severe injury to organs such as the larynx, lung, tympanic membrane, and gastrointestinal tract have been noted in both combat casualty reports [2] and controlled blast tests using live animal subjects [3].

If a live body is exposed to blast overpressure, the events leading to organ injury follow the sequence illustrated in Figure 1. A free field blast produces body loading which produces body distortion and results in local deformation of air-containing organs. Organ stress causes tissue strain which at a critical level will result in injury.

A primary objective in blast overpressure research at JAYCOR is to develop a computer code to analytically predict blast injuries of these organs for both simple and complex blast waves. A major effort in support of this objective is to provide experimental prediction of the injury level of the gastrointestinal (G.I.) tract under various blast conditions, and to gain understanding of the local dynamics at work. To achieve these goals, a series of controlled laboratory procedures was developed to study and directly observe gastrointestinal blast related injury.

By utilizing autologous perfusion, ultrasonic bubble mapping, Hemastix, blood cell count, the probe-in-balloon pressure measurement technique and visual observations, detailed information on blast related injury of the G.I. tract can be quantified. This report summarizes details of these standardized laboratory procedures and shows typical examples of G.I. injury quantification measurements. These techniques can be used to establish the correlation between various blast dynamics parameters and G.I. blast injury. The correlations can then be used to further the development of an analytical model for prediction of G.I. blast injury.

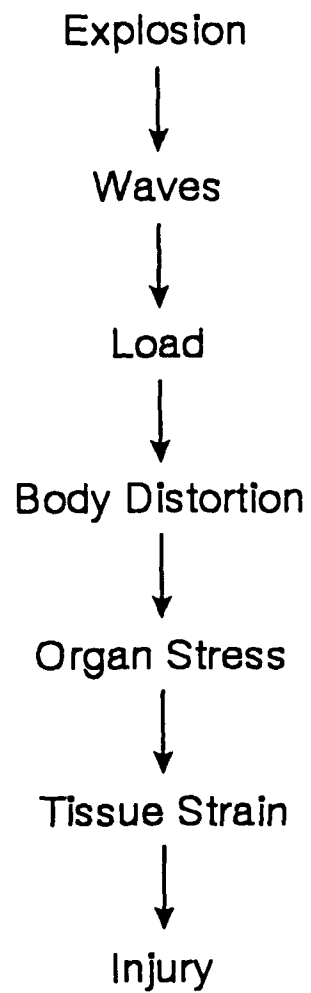


Figure 1. Common mechanical sequence of injury.

2. POWDER ACTUATED BLAST OVERPRESSURE TEST CHAMBER

Field tests showed that a blast wave changed markedly as it passed through the body of a live animal (see Fig. 2). To properly study the local mechanics of gastrointestinal injury, a method for laboratory simulation of intra-abdominal blast signal was needed. By utilizing a commercially available powder actuated impactor, a G.I. blast overpressure test chamber was designed and fabricated.

Figure 3 is a schematic of the basic system components: impactor, piston, diaphragm and chamber. The powder actuated impactor uses 22 caliber blanks to produce an explosive pressure wave which impinges on the piston assembly. The piston assembly delivers an impulse to the flexible diaphragm producing a pressure wave in the fluid filled chamber. The system was specifically designed to produce a signal with characteristics similar to previously measured intra-abdominal blast signals. Typical chamber and sample bubble reaction pressure signals are shown in Figure 4.

The complete blast overpressure test system, shown in Figure 5a, incorporates a recirculating heater-pump to maintain the chamber fluid at the animal body temperature. The lid of the chamber is clear Plexiglas to allow high speed filming of the test organs during blast exposure.

The chamber pressure is adjustable in both magnitude and A-duration through the use of different size powder loads (Fig. 5b). Calibration tests have attained repeatable pressure peak values in the range 15 to 850 kPa. This allows the study of injury mechanism for each different organ section.

ABDOMINAL CROSS-SECTION AT SECOND LUMBAR VERTEBRA

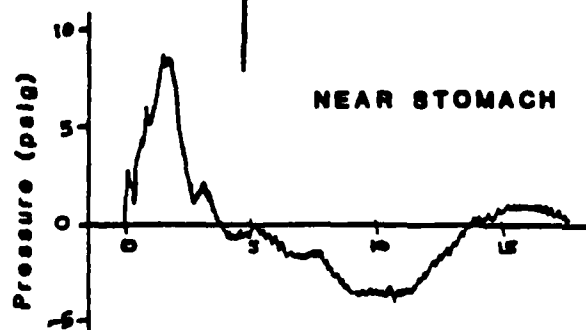
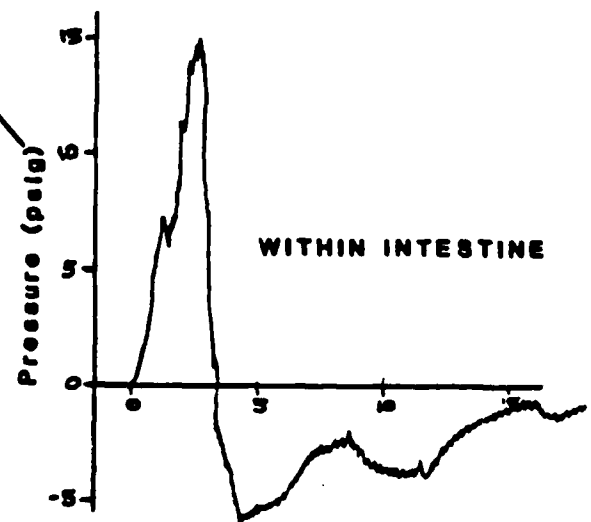
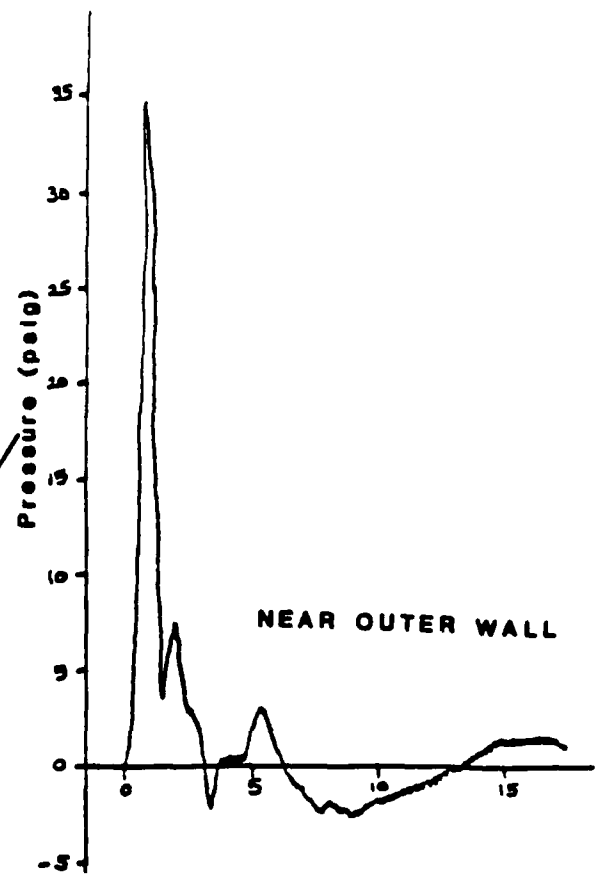
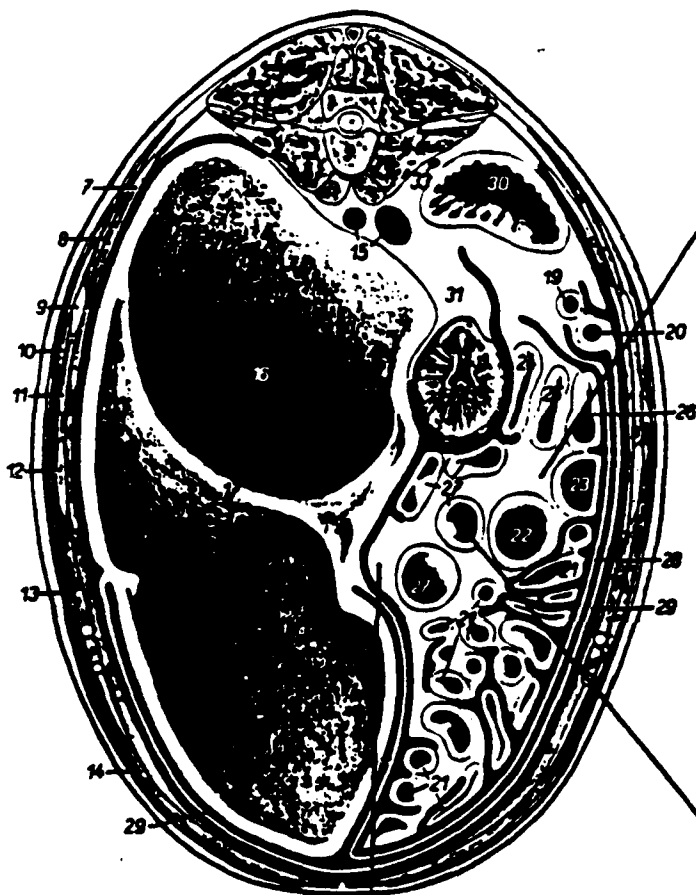


Figure 2. Typical sheep loading signals.

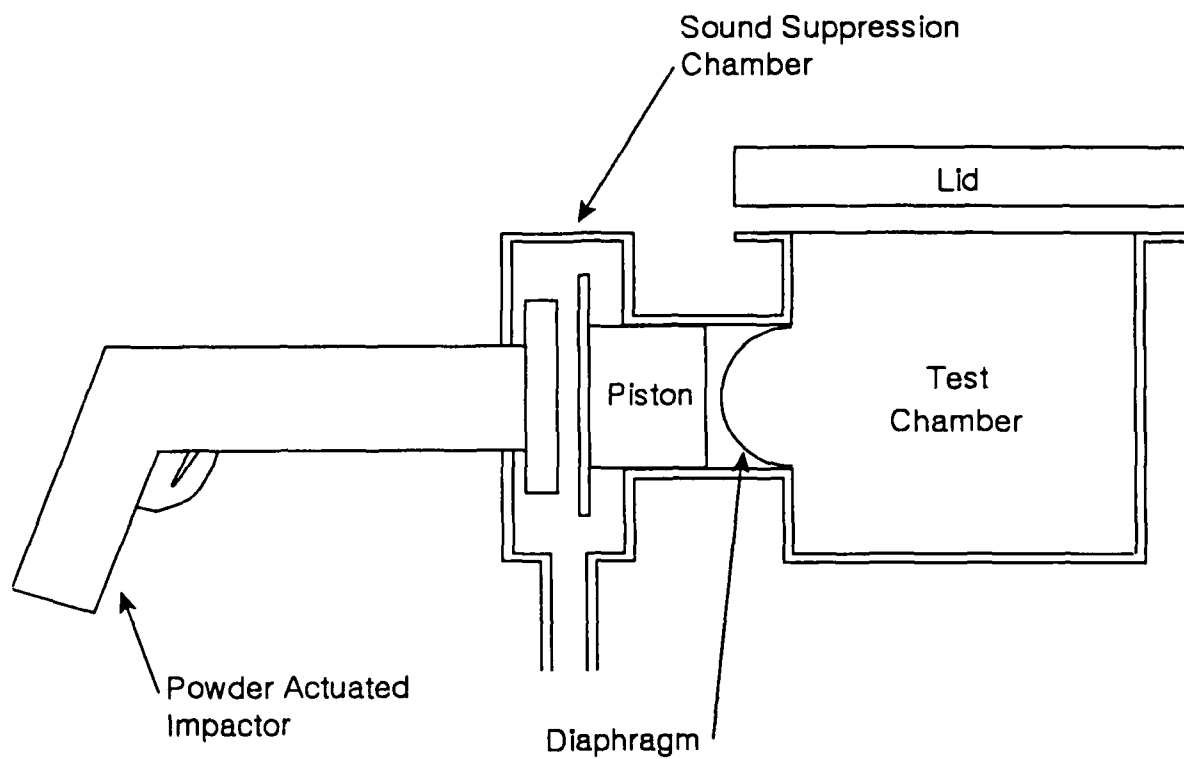
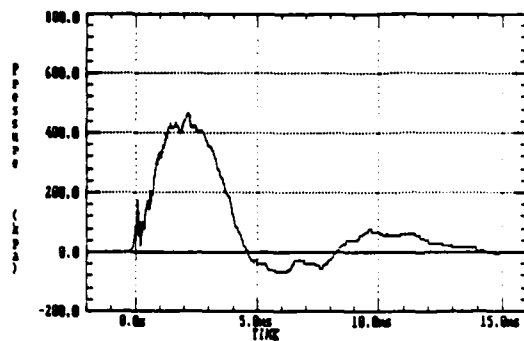
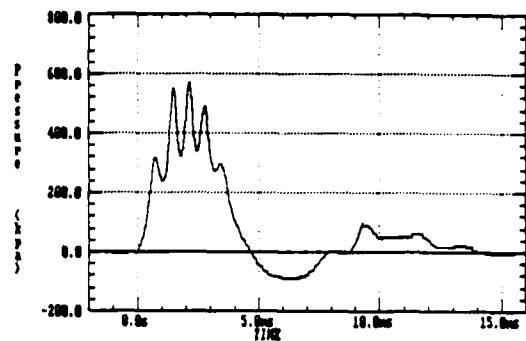


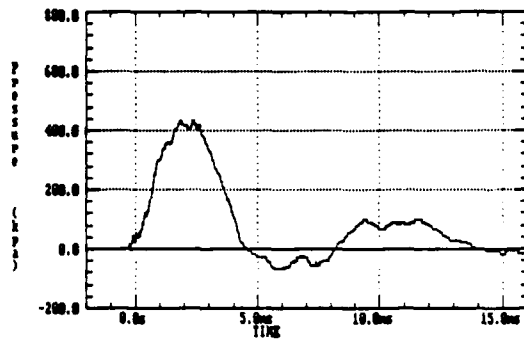
Figure 3. Schematic of blast overpressure basic system components.



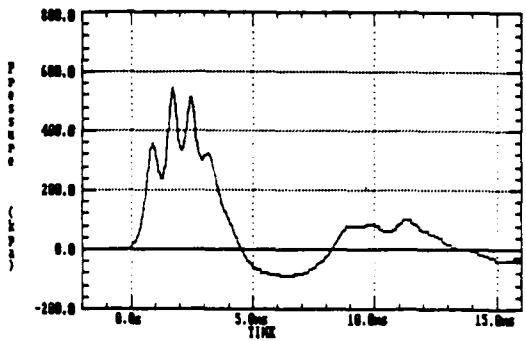
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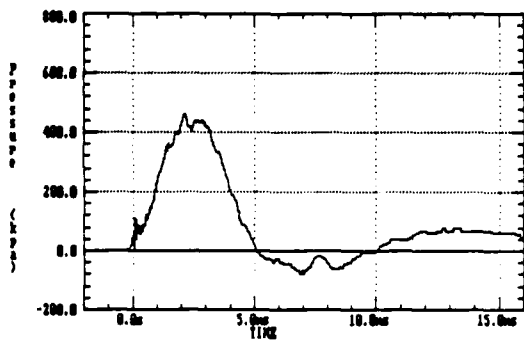
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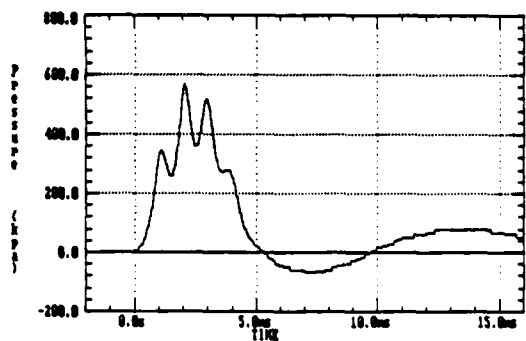
432 kPa



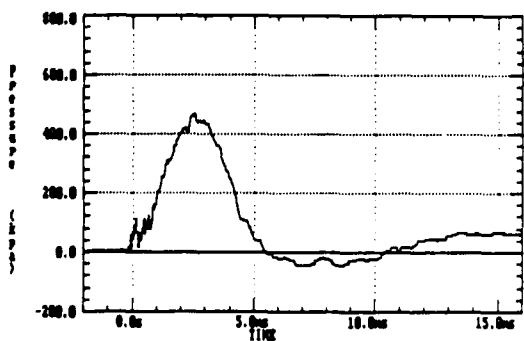
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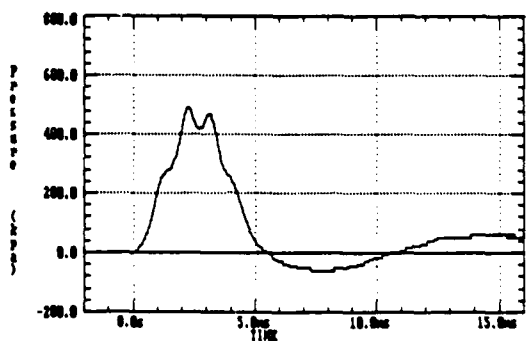
461 kPa



10 cc



461 kPa
P_{CHAMBER}



15 cc
P_{BUBBLE}

Figure 4. Examples of repeatable pressure chamber loading signals and typical bubble responses for 2-15 cc bubbles.

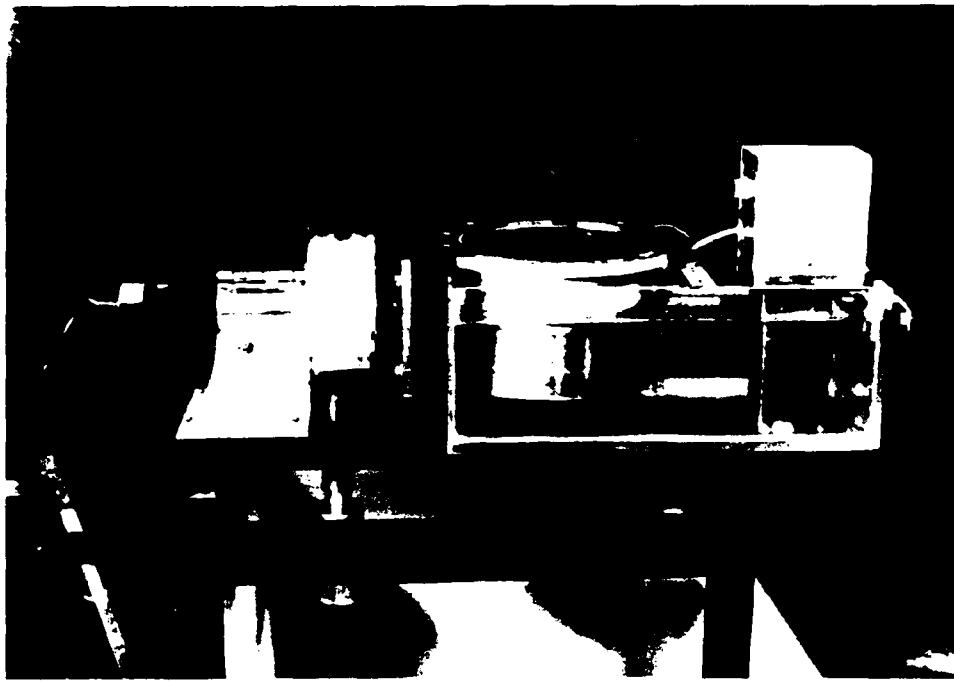


Figure 5a. Laboratory blast loading system: impactor, test chamber and constant temperature bath.

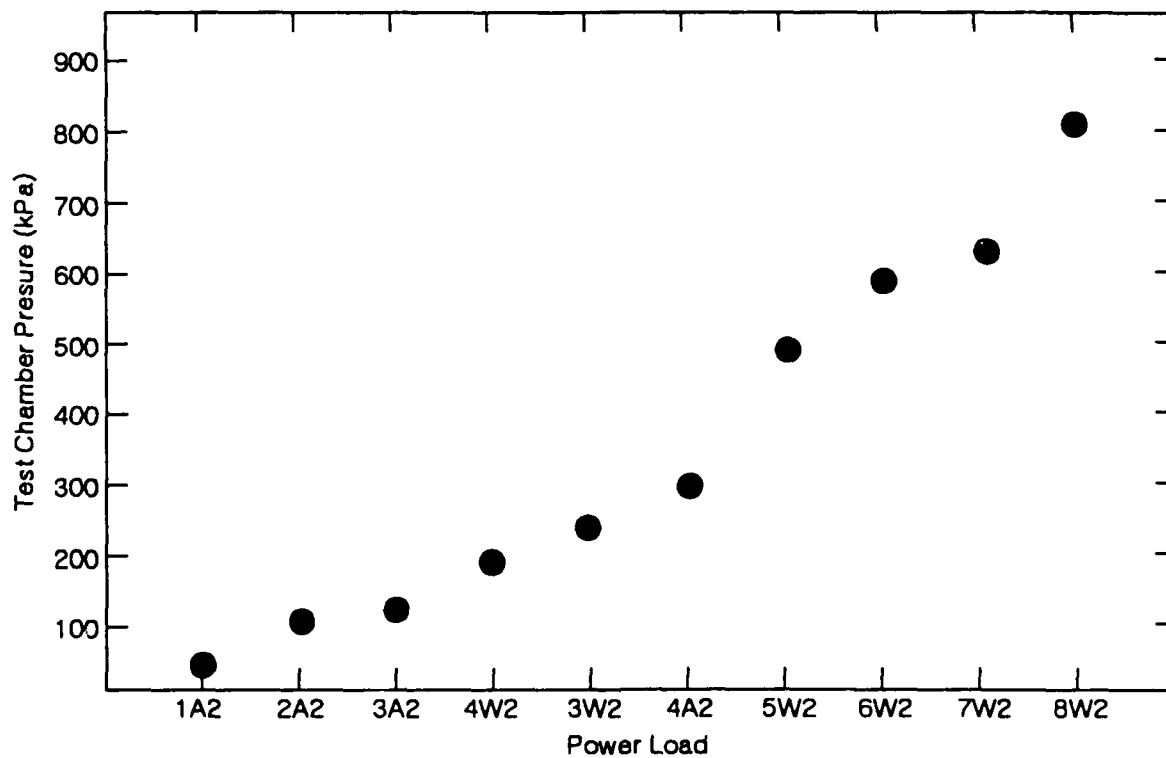


Figure 5b. Test chamber pressure versus power load.

3. ISOLATED GASTROINTESTINAL TRACT AUTOLOGOUS PERFUSION

As stated earlier, air blast with enough intensity and duration can lead to injury of the gas-containing organs of an exposed test animal. The gastrointestinal tract is particularly vulnerable to this type of injury with contusions usually appearing on the large intestine and the stomach [4], because these are the places where gas bubbles are most likely to accumulate. Direct connection between the injury site and the dynamics of gas bubbles can only be established from direct observation and/or quantitative measurements. Therefore, isolated G.I. tract tests are the logical approach for collecting such information.

Using isolated unperfused G.I. tract sections as test samples has two major drawbacks. Because the test section has no blood pressure there is little indication of the occurrence of hemorrhagic injury. Secondly, autolysis usually begins to take place shortly after the blood supply is removed. As a result, little time is available for in-depth study and the test results are of questionable merit.

Conventional isolated organ perfusion techniques also present problems because they usually require a complex blood supply system that includes a pulsating pump, oxygenator, foam suppression equipment, and provisions for prevention of blood cell damage [5]. Since the rabbit's gastrointestinal tract is supplied by only two major arteries and return flow is through one major vein, a perfusion technique was developed to use the test rabbit's own cardiopulmonary system as the autologous blood supply source. The technique allows for the removal of the complete intestinal loop from the abdominal cavity of the rabbit, and maintains its blood circulation through silastic tubing connections of the catheterized cranial and caudal mesenteric arteries with return flow through the portal vein, as shown in Figure 6. The isolated perfused G.I. tract can then be placed in the overpressure test chamber for controlled experiments.

Details of the autologous perfusion procedure are contained in Reference 6. The procedure permits an isolated G.I. tract to be kept viable for several hours, thus allowing more experimental control and direct observance of the progression of post-blast exposure trauma.



Figure 6a. Perfusion loop of isolated G.I. tract. Flow is from abdominal aorta into cranial mesenteric artery, out of portal vein and into caudal vena cava.



Figure 6b. View of perfused isolated G.I. tract showing perfusion tubes passing through test chamber cover plate.

4. PROBE-IN-BALLOON (PIB) UNIT

In support of the study of blast overpressure (BOP) related gastrointestinal (G.I.) injury, a probe-in-balloon (PIB) test unit was designed and tested in the JAYCOR Biomechanics Laboratory. The air-filled unit was shown to accurately represent the amplitude and frequency response of an equivalent gastrointestinal bubble during blast overpressure experiments. It offers a controlled means of measuring the G.I. bubble pressure for in vivo animal field tests and helps establish the direct correlation with G.I. injury. A water-filled PIB could be used to measure the reference abdominal pressure and alleviate some of the uncertainties associated with the current method of probe placement.

BACKGROUND

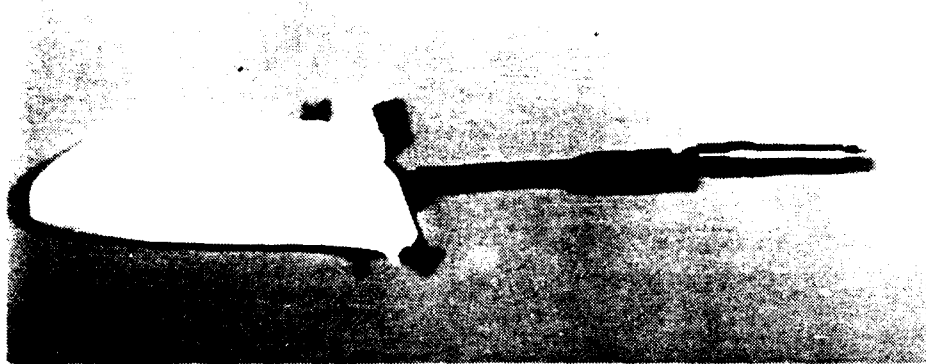
In order to establish the relationship between local stress and injury threshold, however, quantitative information is required. In a laboratory setting, location of the bubble can be controlled and the placement of pressure transducer within the bubble can be accomplished with confidence. Such measurement has been used as a standard procedure for our laboratory tests of perfused G.I. tract.

In field tests, where in vivo animal tests are carried out, keeping the pressure transducer in a free bubble could be difficult. The probe installed during surgical preparation may not stay in the bubble when the body orientation changes, such as during transit to the test location and during field tests. Since the pressure reading at any other location could be markedly different from that in the bubble, the results obtained with uncontrolled bubble-probe relative position could be misleading.

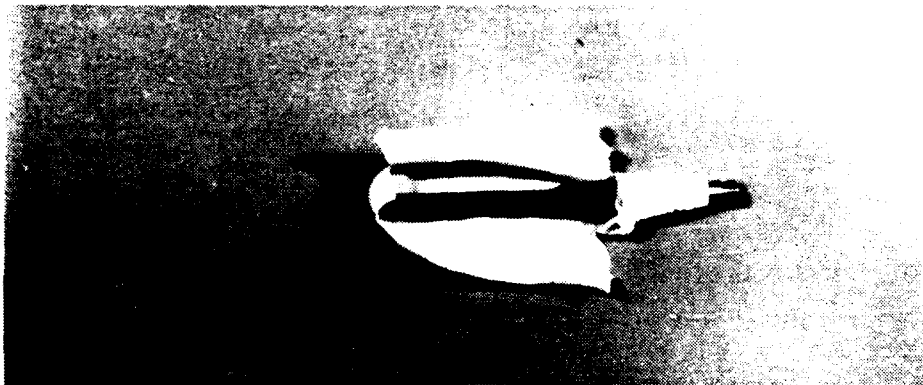
To ensure the probe stays within the bubble and to ensure the test bubble stays coherent during blast exposure, a probe-in-balloon approach was conceived. This approach involves the adoption of a flaccid latex balloon to hold a controlled amount of air to keep the bubble from dispersing, and a slotted support tube to act as a conduit for the bubble pressure cycles during blast exposure.

CALIBRATION

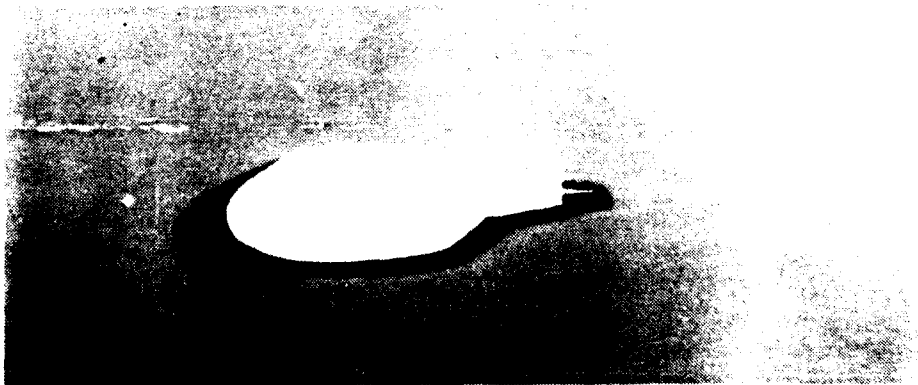
The Probe-in-Balloon (PIB) unit is shown in Figure 7. This device is composed of a Kistler 211B4 pressure transducer mounted inside a limply filled, latex balloon containing a measured volume of air. The cutaway view, Figures 7-a and 7-b, show a plastic, slotted, hollow tube attached to the transducer tip. This tube serves as an air pressure conduit and also allows the device to be more easily placed within a test animal's intestinal contents.



(a)



(b)



(c)

Figure 7. Probe-in-balloon ("PIB") unit.

Because the latex balloon is limply filled to about 70% of its maximum volume, the material is likely to experience little strain during both expansion and compression modes and thus has minimal effect on the bubble pressure signal response. Figure 8 shows the calibration test setup where a PIB unit with a 5 cc air bubble was placed next to a 5 cc free air bubble in a lambskin surrogate. A reference transducer was used to measure the lambskin air bubble pressure. The whole unit was then placed in the test chamber shown in Figure 9 for calibration tests. Typical calibration signals are shown in Figure 10, and the results are summarized in Figure 11. As shown, when the chamber pressure is relatively low, the difference between the peak pressures of the free bubble and the PIB bubble is negligible. However, there is a systematic reduction in amplitude for the PIB signal when the loading pressure is increased. This lower PIB reading could be the result of flow restrictions through the support spine. This difference could be minimized by using a spinal support that has little flow blockage such as one made of wire. Otherwise the difference can be compensated for by direct calibration of the chosen unit.

CONCLUDING REMARKS

The PIB unit when placed in a surrogate G.I. has been shown to truly represent the amplitude and oscillation frequency of an equivalent gastrointestinal bubble. It allows the freedom to control both the bubble location and bubble volume so that their effects on injury thresholds can be more reliably determined.

The PIB unit can also be completely filled with water and used to measure the abdominal pressure under blast loading. Such information is essential in establishing the representative abdominal loading for G.I. injury threshold and determining the connection between laboratory test chamber data and field test results.

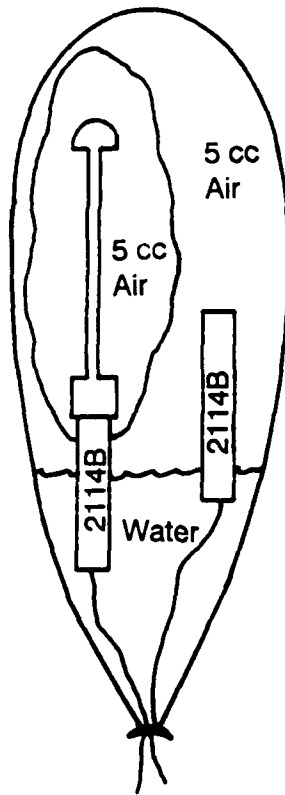
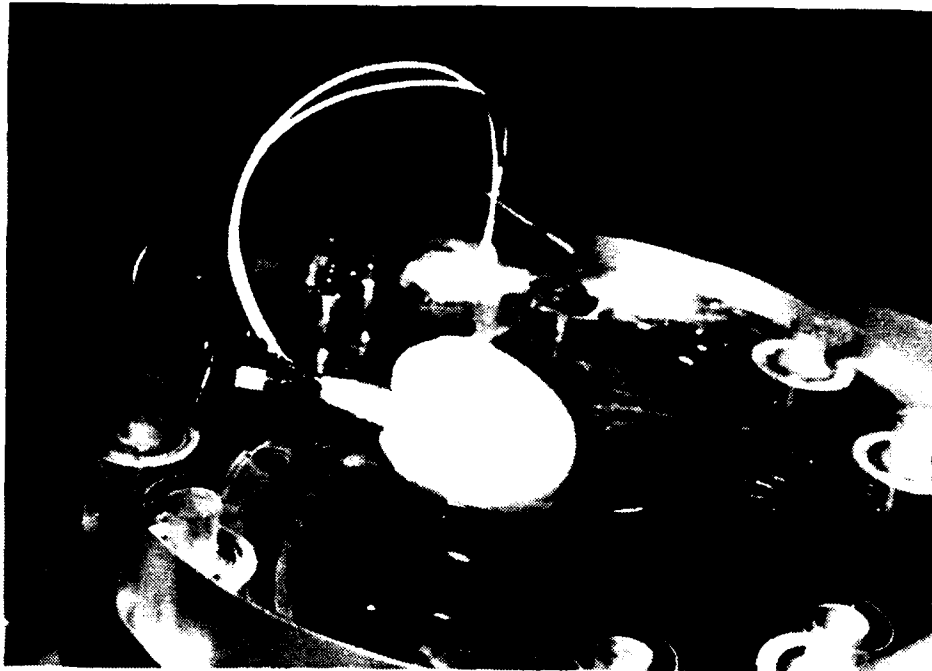


Figure 8. Schematic of initial PIB test setup.

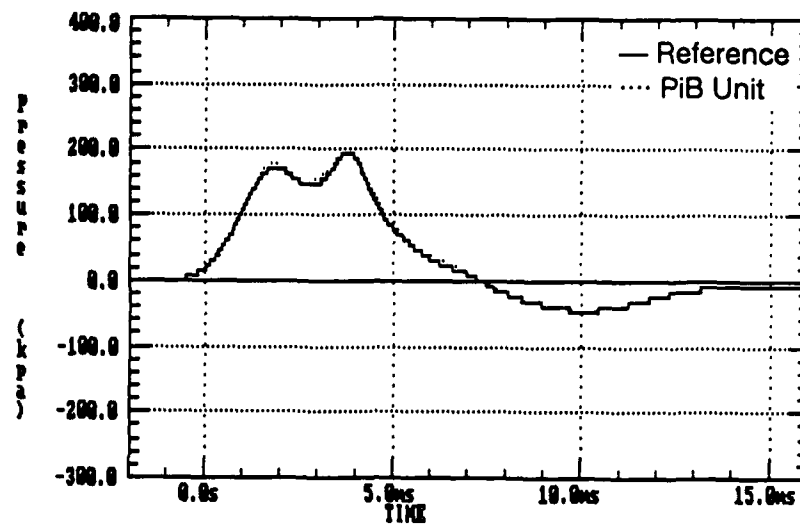


(a) PiB calibration setup and test chamber

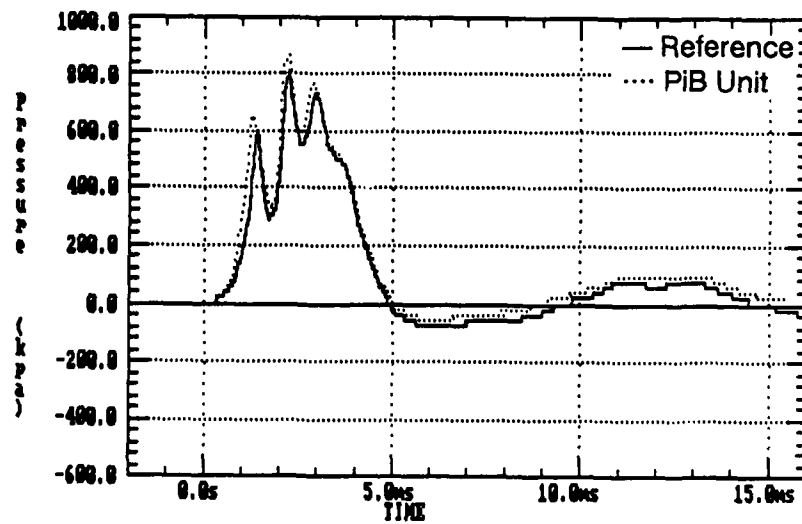


(b) Cutaway view

Figure 9. PiB calibration setup.



(a) 10 psi loading



(b) 120 psi loading

Figure 10. Typical PIB calibration signals.

FREE BUBBLE vs. PiB BUBBLE

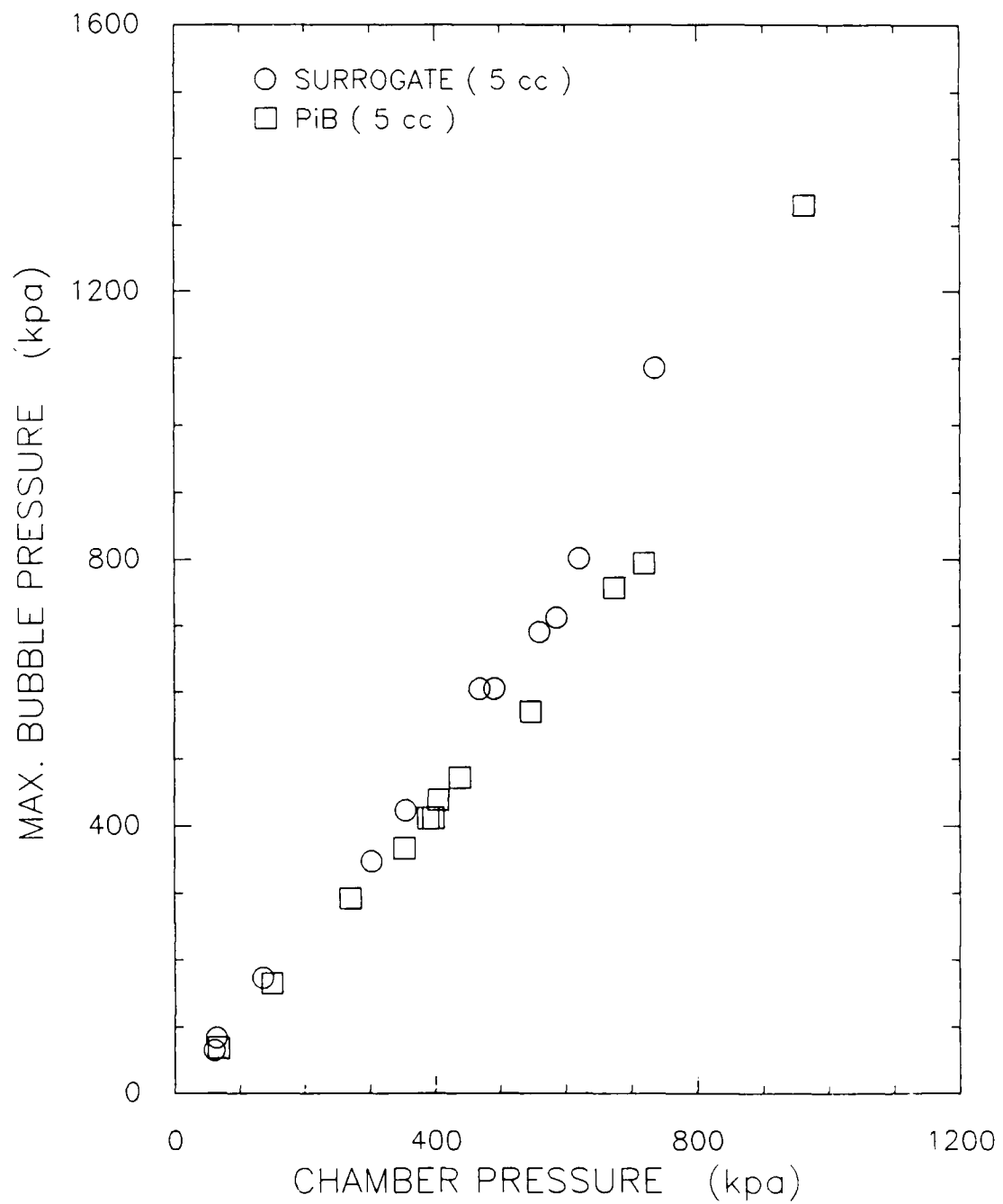


Figure 11. A comparison of maximum bubble internal pressure magnitudes for a 5 cc free bubble in a lambskin surrogate and a 5 cc PiB in a lambskin surrogate.

5. G.I. GAS BUBBLE MAPPING

Prior to blast exposure, bubble mapping of the G.I. tract is accomplished using an ultrasonic scanning instrument. This determines the location and approximate size of any naturally occurring G.I. gas bubbles.

A NOVA 201A ultrasonic thickness gauge, shown in Figure 12, was originally used to measure the thickness of the intestine wall. Since the instrument measures thickness by using the traveling time of Megahertz frequency pulses between transmitted and echoed-back signals from a substrate to an air interface, it can also be used to indicate the presence of air bubbles. For example, the D2 sensor has a 12 MHz frequency and a resolution of 0.0001 inch for measurement of intestine wall thickness. A gas bubble directly underneath the point of measurement would provide an interface indicating a thin walled membrane, thus a positive indication of a gas bubble just underneath the wall. If a full cross-sectional thickness of the G.I. section is displayed, it would indicate no gas bubble presence. An intermediate thickness value would register a fecal entrained gas bubble. By traversing along the whole length of the gastrointestinal tract, marking the length and location of gas bubbles, a gas bubble distribution map for the test animal is established. This information serves as a benchmark for determining injury correlations. If no natural gas bubble is found in a desired section, an air bubble may be injected or a PIB (Section 4) may be placed to gather pressure data for a specific size bubble. This option of bubble placement further reduces the total number of test animals needed to gather a specific data base.



Figure 12. NOVA 201A digital ultrasonic thickness gauge used to map gastrointestinal bubbles.

6. G.I. TRACT BLAST INJURY DETERMINATION PROCEDURE

One of the goals of G.I. blast injury studies is to establish *quantitative* correlates of injury to the gastrointestinal tract that can be compared with computational models and field data. A second goal is to detect the threshold of hemorrhaging so that relative vulnerability of each type of G.I. section can be established.

Figure 13 shows an example of colon serosal blast injury. This type of injury is quantified through direct physical measurement of the affected area; such as the intestine diameter, area of contusion, wall thickness, gas bubble volume, bubble internal pressure and intestine fill condition. The intestine diameter and area of contusions can be directly scaled and the wall thickness can be determined using the ultrasonic thickness gauge (Section 5). Calibration tests have shown that the gas bubble volume can be accurately determined by syringe withdrawal and the bubble internal pressure during blast can be measured using a PIB (Section 4). The degree of intestine fill can be derived by tying off the injured test section with ligatures and injecting measured amounts of saline solution into the test section until it is full. Then the test section's total fecal slurry volume can be withdrawn and compared with the measured volume of fill saline. This ratio can be used as a "fill value." All of these measured parameters can then be correlated with serosal injury and blast overpressure level to determine threshold dynamics.

Mucosal injury usually takes place at a lower blast level. Although the injured section parameters can be measured using the same techniques as described for serosal injury, quantifying the mucosal injury level is different. Since this type of injury involves bleeding into the fecal contents of the G.I. tract (see Fig. 14), an indication of mucosal injury is the testing of the fecal slurry for the presence of blood before and after blast exposure. Two methods of quantifying mucosal type G.I. blast injury in this manner are the use of Hemastix and blood cell counting.

Hemastix is a commercially available product for testing the presence of hemoglobin in urine at levels as low as 0.062 mg/dl. Our calibration tests have found that Hemastix can be used as an effective indicator of G.I. mucosal injury. Although proper dilution factors must be used to insure against false positives due to microbial peroxidase in the fecal slurry, a positive indication of mucosal bleeding has been found at dilution levels as low as 2 ppm. Hemastix used to test diluted fecal slurry samples taken before and after blast exposure give an indication through color change that mucosal bleeding has occurred (Fig. 15). Though the degree of color change indicates a relative concentration of hemoglobin, a microscopic blood cell count of the diluted before and after exposure fecal samples allows a more accurate quantification of the mucosal injury for data comparisons (Fig. 16).

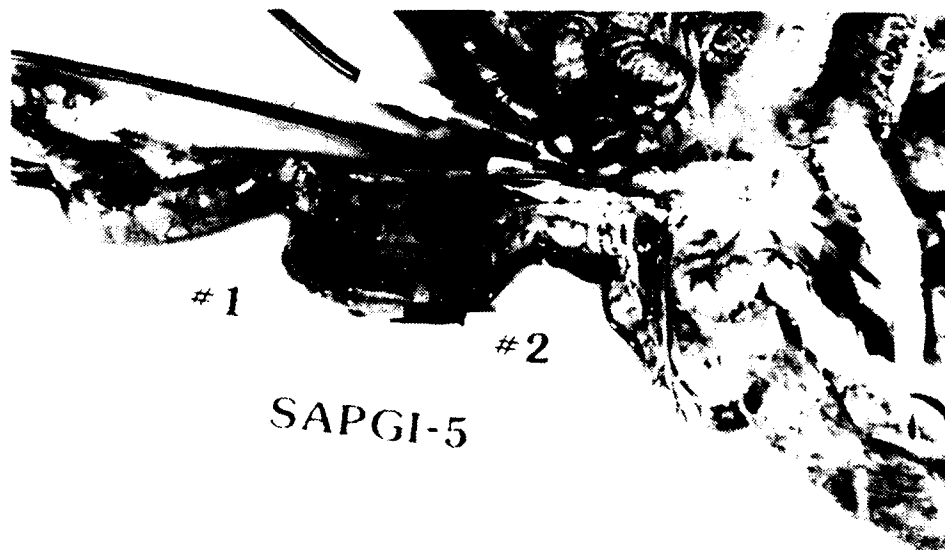
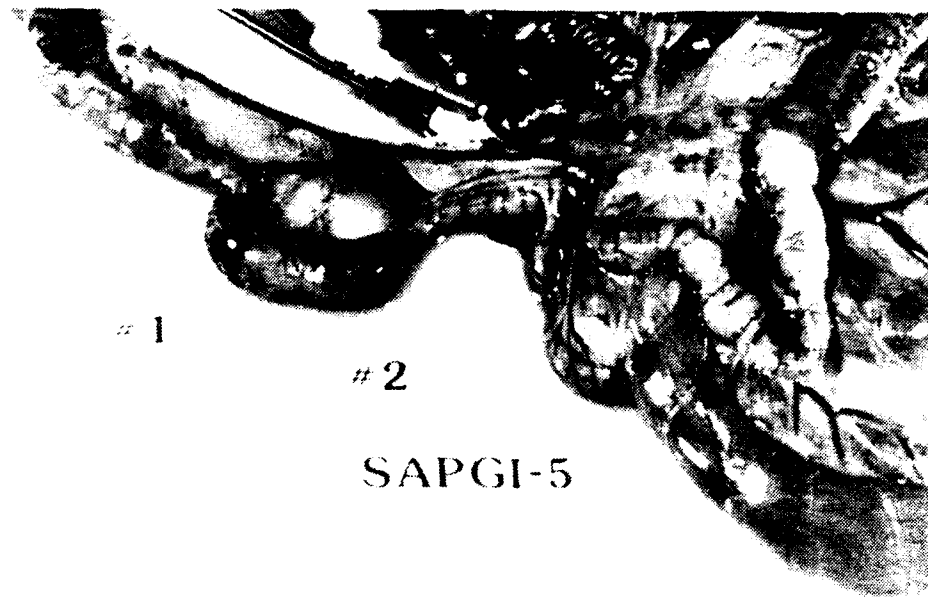


Figure 13. An example of natural state colon serosal injury.

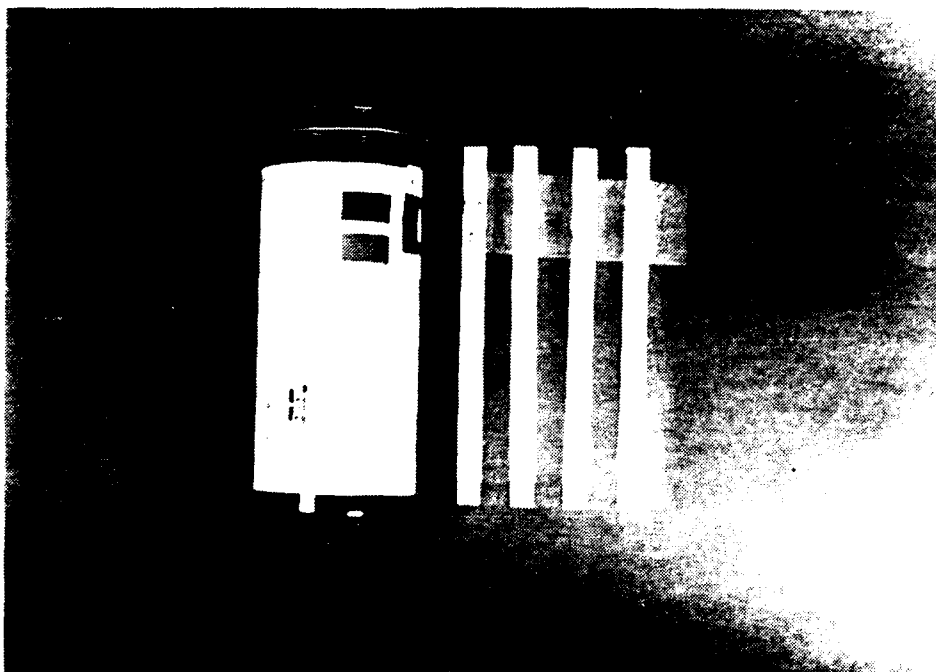


#2



#2

Figure 14. An example of natural state caecum mucosal injury.

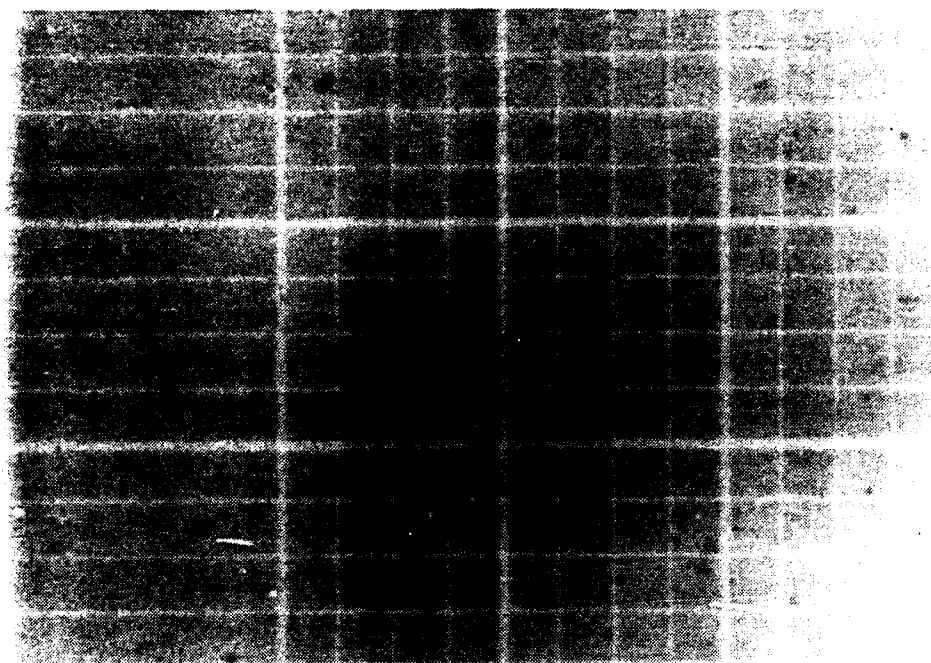


(a) Hemastix strips

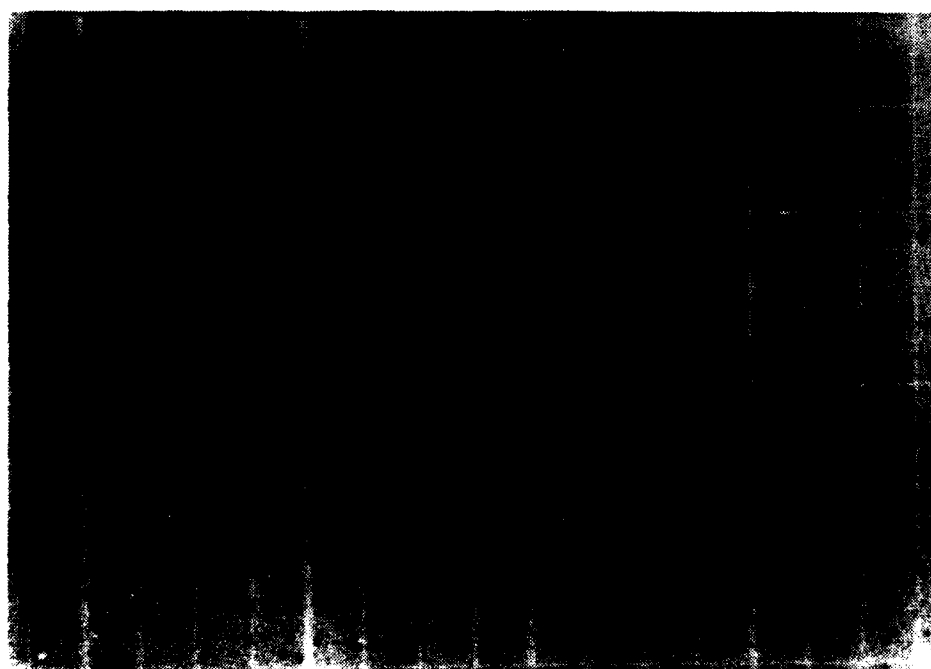
BEFORE EXPOSURE AFTER EXPOSURE

(b) Actual test results

Figure 15. Hemastix test strip for mucosal injury indication.



(a) Before exposure



(b) After exposure

Figure 16. Blood cell count as mucosal injury indicator.

7. GENERAL PROCEDURES FOR GASTROINTESTINAL INJURY DETERMINATION

The injury quantification techniques have been standardized into the routine procedures summarized below.

7.1 BLAST OVERPRESSURE EXPOSURE

Prior to exposure, isolate and perfuse the gastrointestinal tract. Install PIB and/or inject air bubbles at test locations. Take fecal slurry sample before exposure. Expose the isolated, perfused tract to blast in the pressure chamber. After the exposure, wait 30 minutes to allow contusions to develop.

7.2 PHYSICAL MEASUREMENTS OF SEROSAL INJURIES

Clamp off the perfusion tubes, euthanize the animal. Remove the test section from the chamber and visually inspect the G.I. tract for injury. Photograph the G.I. tract. Perfuse the isolated arterial system with saline solution through the cranial mesenteric artery to replace all non-hemorrhagic blood. Photograph the purged G.I. tract and any serosal injuries. Perform physical measurements of injury section geometry and determine injury site local gas volumes. Perform mucosal injury determination.

7.3 MUCOSAL INJURIES DETERMINATION

Dilute pre-blast reference fecal samples in saline at 1:100, or until no microbial-induced positive reaction is seen on the Hemastix test strips. If serosal injury is not shown gather post-blast fecal sample at the bubble sites. Dilute fecal sample to the same concentration as pre-blast references, and test with Hemastix for immediate injury indication. If Hemastix indicates positive hemoglobin presence, then perform microscopic blood cell count on the pre- and post-blast diluted samples. Perform physical measurements as needed for injury correlations.

8. CONCLUSIONS

Through the use of various laboratory techniques including gastrointestinal autologous perfusion, ultrasonic gas bubble mapping, fecal hemoglobin sampling and blood cell counting, a series of standardized G.I. blast injury procedures were developed. These techniques can be used to establish quantitative determinations of injury to the gastrointestinal tract that can be compared with computational models and field data. The procedures allow direct measurement of G.I. gas bubble dynamic pressure, bubble volumes, and injured section geometric characteristics. Of special importance is the ability to detect the first signs of hemorrhaging and therefore isolate the effects on G.I. injury thresholds of a specific blast dynamics parameter.

Highlights for each procedure are given below.

- The powder actuated test chamber can deliver an intra-abdominal type blast signal in a range between 15 and 850 kPa.
- The autologous perfusion procedure can keep an isolated gastrointestinal tract viable for several hours of controlled, observable testing.
- The probe-in balloon (PIB) unit can be used to quantify G.I. bubble internal pressure time histories during blast overpressure exposure.
- Procedures have been established to measure specific injury parameters and to quantify serosal and mucosal G.I. injury using a blood cell count and a hemoglobin indicator.

These laboratory procedures can be used to investigate the local mechanism of blast related gastrointestinal injury. This will facilitate the development of an analytical model to predict G.I. injury and establish a DRC for blast overpressure exposure.

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