Mechanical flexural *ex vivo* study of osteotomized swine femurs stabilized with two types of polyamide 12 rods

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ABSTRACT: *Long bone fractures are commonly in surgery routine and several bone imobilization techniques are currently available. Technological progress has enabled to use low cost materials in surgical procedures. Thus, the aim of this study was to evaluate the applicability of polyamide 12 rods, solid and hollow in swine femurs, comparing them through flexion strength. This study had as second aim to fix the locking errors, commom place in interlocking nails, once polyamide 12 allows perforation in any direction by orthopaedic screw. Six groups were used: G1 - eight whole swine femurs; G2 - eight whole swine femurs with drilled medullary canal; G3 - two solid polyamide 12 rods; G4 - two hollow polyamide 12 rods; G5 - eight osteotomized drilled swine femurs with a solid polyamide 12 rod implanted in the medullary canal and locked by four 316L stainless steel screws; and G6 - similar to G5 but using hollow rods instead of solid ones. No significant differences were observed for the modulus of rupture between solid and hollow rods, demonstrating that both rods had similar performances. These results led to the speculation that the addition of other polymers to the hollow rods could increase their strength and thus the bone-implant system. Furthermore, the comparison between G1, G5 and G6 could be analyzed using the finite element method in future. New polymeric materials may be developed based on the data from this study, strengthening the bone-implant system and making possible screws to be placed in any direction, nullifying the detrimental forces on the fracture site.*

Key words: *biomechanical test, implants, interlocking nail, prototyping, laser sintering.*

Estudo mecânico de flexão, *ex-vivo*, de fêmures suínos osteotomizados, estabilizados com dois tipos de haste de poliamida 12

RESUMO: *Fraturas em ossos longos são comumente encontradas na rotina cirúrgica e várias técnicas de imobilização óssea estão disponíveis. Com o avanço tecnológico, tornou-se viável utilizar materiais de baixo custo nos procedimentos, portanto esse estudo objetivou avaliar a aplicabilidade de hastes de poliamida 12, sólidas e vazadas, implantadas em fêmures suínos, comparando-as segundo as forças de flexão e aos erros de bloqueio, corriqueiros nesse implante, uma vez que a poliamida 12 permite sua perfuração em qualquer direção por meio de parafusos ortopédicos. Seis grupos foram usados: G1 - oito fêmures suínos íntegros; G2 - oito fêmures suínos, fresados intramedularmente; G3 - duas hastes maciças de poliamida 12; G4 - duas hastes vazadas de poliamida 12; G5 - oito fêmures suínos osteotomizados e fresados, com haste de poliamida 12 maciça implantada no canal medular e bloqueada com quatro parafusos de aço inoxidável 316L e G6 - diferente de G5 apenas por utilizar hastes vazadas. Não foram observadas diferenças significativas no módulo de ruptura entre hastes sólidas e vazadas, demonstrando que ambas apresentaram o mesmo desempenho. Estes resultados levaram à especulação de que adicionar outros polímeros às hastes vazadas aumentaria sua força e, portanto, do sistema osso-implante. Além disso, a comparação entre G1, G5 e G6 poderia no futuro ser analisada utilizando o método dos elementos finitos. Novos polímeros podem ser desenvolvidos baseando-se nos dados deste estudo, reforçando o sistema osso-implante e também possibilitando o uso de perfurações para o bloqueio no transoperatório em qualquer direção, anulando as forças deletérias atuantes no sítio de fratura.*

Palavras-chave: *ensaio biomecânico, implantes, haste bloqueada, prototipagem, sinterização a laser.*

INTRODUCTION

Long bone fracture is one of the main orthopaedic conditions in Veterinary Medicine, with femur fractures representing 20 to 25% of all long bone fractures in cats and dogs. Unfortunately, there is currently no stabilization method that can be applied to all types of long bone fractures but every procedure has its pros and cons (DIAS, 2006). However, the Interlocking Nail technique described

by Modney is a stabilization method that combines the biomechanical advantages of intramedullary rods to the stiffness of plates and screws and their ability to block the forces on the fracture site (GUPTA, 2001). Forces acting on the fracture site are minimized by fixing the locked rod to the bones with screws. Thus, interlocked rods are considered to be biomechanically superior to intramedullary rods, plates or external fixators (HORSTMAN et al., 2004).

There is a call for rods made of light resistant low cost materials that are biocompatible with the bone tissue (VAN Der ELST et al., 1999). In this context, polymeric materials have been continuously discovered, tested and incorporated into surgical procedures, both in human and animal medicine (SPADETO JUNIOR et al., 2011). According to WIEBECK & HARADA (2005), polymeric materials can be manufactured by Selective Laser Sintering (SLS). Furthermore, polyamides are of low cost, easy acquisition, versatile and can be moulded into various shapes and sizes. The polyamide 12 stands out in the manufacturing of surgical materials due to its low absorption of water, thus not requiring drying prior to processing; low absorption of body fluids; therefore, minimizing the chances of contamination and for its easiness of perforation during the surgical procedure.

The aim of this study was to analyse the applicability of solid and hollow polyamide 12 locked rods in osteotomized swine femurs, as well as compare the differences in the bending force (flexion) of both rods through mechanical tests.

MATERIALS AND METHODS

Thirty-two swine femurs obtained at a commercial abattoir were radiographed and selected for their structural integrity. Femurs of homogeneous weight, size and shape were selected. The bones were dissected and cryopreserved in a conventional freezer at -20 to -25ºC until needed, as recommended by GALUPPO et al. (2002) and DALLABRIDA et al. (2005). The polyamide 12 rods, with 15mm diameter and 150mm in length, were manufactured by Selective Laser Sintering (prototyping) at the Centro de Tecnologia da Informação Renato Archer, Divisão de Tecnologia Tridimensional, Campinas – SP, Brazil. They were produced in solid and hollow forms. The hollow rods were longitudinally perforated in the centre and had an internal diameter of 1/3 of the external one.

For the mechanical tests, the bodies of proof (femurs and rods) were distributed into six experimental groups: G1 - eight whole femurs, not osteotomized or drilled; G2 - eight whole femurs, not osteotomized but with drilled medullary canal; G3 - two solid polyamide 12 rods (Figure 1A); G4 - two hollow polyamide 12 rods (Figure 1B and 1C); G5 - eight osteotomized drilled femurs, with a solid polyamide 12 rod implanted in the medullary canal and interlocked by four 316L stainless steel orthopaedic screws; and G6 - different from G5 due to the use of hollow rods instead of solid ones.

The femurs were removed from the freezer and submerged in filtered water, at room temperature, for 24 hours. The body of proof of all groups, after their respective preparations, were submerged in a similar manner for 24 hours to ensure that the materials were saturated due to their hygroscopic properties. Once the preparation was over, the flexion tests were applied according to the ASTM D790-00 guidelines – "Standard Test Methods for Flexural Properties of Unreinforced and Reinforced Plastics and Electrical Insulating Materials". An Emic® DL10000 universal test machine, with 100kN load cell and speed of 2mm min^{-1} was used for the three-point flexion tests, with the third loading pin being placed equidistant to the two supporting pins (*span*). The *span* was chosen to prevent the body of proof from sliding and so that it's central region was free for test. A distance of 100mm was adopted for all groups. Tests were undertaken at the Laboratório de Engenharia Mecânica da Universidade de Franca (UNIFRAN), Franca-SP, Brazil.

Osteotomy was performed in groups G5 and G6 using an orthopaedic saw and creating a transversal fracture in the middle of the femoral diaphysis. The medullar cavity was drilled continuously and progressively with 5 to 15nm manual drills, on both osteotomized bone fragments. Subsequently, the solid (G5) and hollow (G6) rods were manually inserted into the drilled cavity so as to ensure an anatomical reduction of the fracture. As they were of the same diameter as the drilled hole, the rods fitted tightly, with no room for movement. Four points (two in each bone fragment), equidistant from each other and from the osteotomy line, were selected for insertion of the screws. A pneumatic orthopaedic drill containing a 2.5mm diameter orthopaedic drill piece was used to make the screw holes, which went through the first cortical bone, the rod and the opposite cortical bone. The bone-implant system was locked by tightening the 3.5mm diameter 316L stainless steel orthopaedic screws of adequate length. Drilling and positioning of screws followed the lateromedial disposition, parallel to the osteotomy line as described by DUELAND et al. (1996) and DUHAUTOIS (2003).
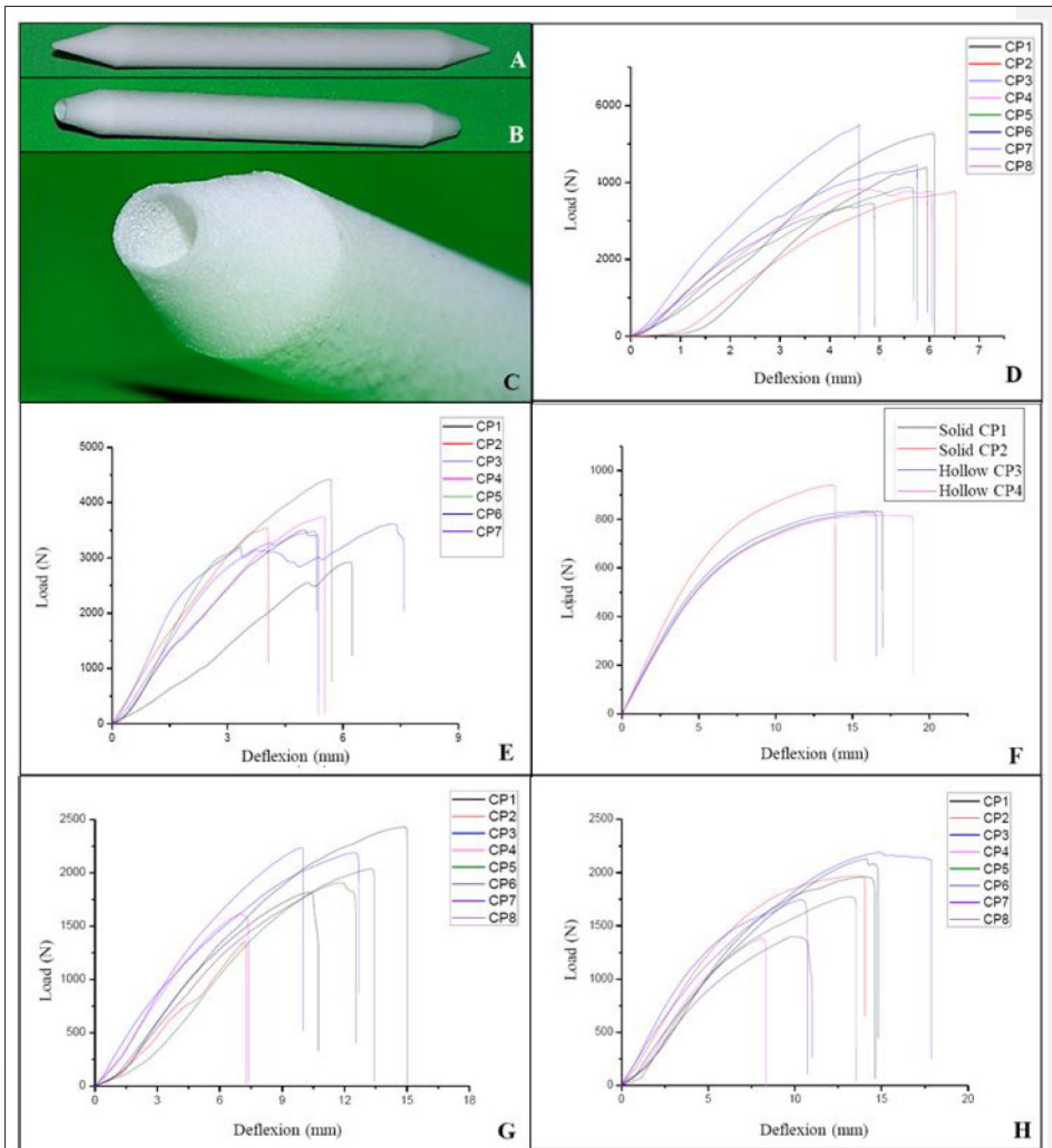
Figure 1 - Photographic images of polyamide 12 rods and the load deflection curves of the experimental groups. A) Solid polyamide 12 rod from group G3; B and C) Hollow polyamide 12 rod from group G4; D) Load deflexion curves of intact femurs not subjected to drilling or osteotomy, group G1; E) Load deflexion curves of intact femurs subjected to drilling but not osteotomy, group G2; F) Load deflexion curves of the solid and hollow rods, G3 (n=2) and G4 (n=2), respectively; G) Load deflexion curves of group G5, bone-implant system with solid polyamide 12 intramedullary rod interlocked with 2316L stainless steel orthopaedic screws in each osteotomized bone fragment; H) Load deflexion curves of group G6, bone-implant system with hollow polyamide 12 intramedullary rod interlocked with 2316L stainless steel orthopaedic screws in each osteotomized bone fragment.

As the drilling of the femoral intramedullary canal was necessary for the insertion of the rods in groups G5 and G6, it was proposed that a test should be performed to verify the fragility of the bone following the surgical procedure. Thus, non-osteotomized swine femurs with (G2, Figure 1E) and without drilling (G1, Figure 1D) were used to test whether there was a weakening of the bone after

the procedure, which could in turn weaken the bone-implant system (G5 and G6).

Isolated flexion tests were carried out on solid (G3) and hollow (G4) polyamide 12 rods (Figure 1F) in order to verify if alterations in their geometry led to significant changes in their mechanical properties, modulus of elasticity and modulus of rupture during flexion. Lastly, tests were performed on the bone-implant system containing solid (G5) and hollow (G6) rods (Figure 1G and 1H) to compare their mechanical properties with those of whole bones. Flexural modulus for the solid rods (G3) was determined by equation 1: $E_M = (4 \times L^3 \times m)/(3 \times \pi \times D^4)$. For G4 (hollow rods) equation 2 was used: $E_V = (4 \times L^3 \times m) / [3 \times \pi \times (D^4 - d^4)]$, where "L" corresponds to the distance between supports (span); "m" the gradient of the initial straight line portion of the load deflexion curve; "d" the internal diameter of the body of proof; and "D" the diameter of the body of proof (GARCIA et al., 2012).

Tension at rupture during flexion was calculated using equation 3: $\sigma_{max} = M_{f\,Max}/W_f$, where M_{fMax} corresponds to the maximum bending moment and W_f to the modulus of resilience to flexion. Maximum bending moment occurred half way between the fixed support points of the *span* machine and; therefore, equation 4 was used: $Mf_{Max} = (F \times L)/4$, where "F" corresponds to the load applied in the test and "L" the distance between the support points. Equation 5 was used to determine the modulus of resilience to flexion for solid circular section with D diameter: $Wf = \pi \times (D^3/32)$. For circular hollow section, with external diameter "D" and internal diameter "d", equation 6 was used: $Wf = \pi \times (D^4 - d^4)/32 \times D$ (GARCIA et al., 2012).

After the tests, the internal and external diameters of the ruptured transversal section from bones and rods were measured in order to calculate the geometric parameters of each sample so that the mechanical properties could be determined. The Student t-test was used to compare the means from the modulus of elasticity and tension at rupture between G1 x G2, G3 x G4 and G5 x G6. Significance was considered at 5% (P<0.05).

RESULTS AND DISCUSSION

Swine femurs were chosen for this study for the easiness and speed to which they could be obtained, especially as standardized sizes had to be used. Furthermore, obtaining the same material from dog could have proven to be complicated and comparison with the literature would have been limited as most studies have used large animal bones for analysis (SPADETO JUNIOR et al., 2011). Polyamide rods were inserted by the normograde way (G5 and G6), which is the most commonly used procedure for canine femur fractures when conventional rods are used, as recommended by SPADETO JUNIOR et al. (2011).

Contrary to SPADETO JUNIOR et al. (2011), who used electric drills and drill bits, the bones in this study were drilled manually. Nevertheless, the present study and those mentioned above are in agreement with FAIRBANK et al. (1995), who recommend the removal of all soft tissue to maximize the contact surface between the cortical bone and the implant, thus strengthening the bodies of proof.

Flexion tests for rigid and semi-rigid materials often determine the modulus of elasticity and resilience to flexion; the former being used as the most important criteria when evaluating the stiffness of polymeric materials and the later being equal to the maximum tension in the external fibres of the body of proof of a polymer at the time of rupture (GARCIA et al., 2012). In the present study, the flexion test was applied to all study groups. Similarly to SPADETO JUNIOR et al. (2011), whole non-drilled bones were used as controls (G1); however, this study also used as controls whole bones that had been drilled but not osteostomized (G2) in order to analyse a potential weakening of the body of proof due to drilling.

No significant difference (P=0.30) was observed in the flexion tests between G1 and G2 (Figure 1D and 1E, Table 1), demonstrating that the drilling did not affect the resilience of the body of proof. Thus, G1 (intact bone) was used as the control for the comparison analysis between the groups (G3 to G6). Results from the body of proof 8 (BP8) from G2 were disregarded as its values differed extremely from the rest. Results for the modulus of elasticity under flexion of the polyamide 12 rods manufactured by SLS (G3 and G4) can be reported on figure 1F and table 2. The data obtained by ASTM D790 was the same as that provided by the distributer (3D Systems).

Sintered polyamide 12 was chosen for this study based on the reports from several other studies that demonstrated its superiority to other materials, such as polyacetal and polypropylene. Furthermore, when compared to previous studies, the manufacturing method used in this experimental model is still unprecedented (SPADETO JUNIOR et al., 2010; SPADETO JUNIOR et al., 2011).

No significant difference (P=0.046) was observed between the mechanical properties of G3 (solid rods) and G4 (hollow rods), which is probably

Table 1 - Flexion test results (Modulus of elasticity and rupture) of G1 (eight whole swine femurs, not osteotomized or drilled) and G2 (seven whole swine femurs, not osteotomized but with a 15mm diameter drilled medullary canal).

		BP1	BP2	BP3	BP4	BP5	BP6	BP7	BP8	Mean	Standard Deviation
Modulus of Elasticity (MPa) – MOE	G1[1]	2014.59	1169.81	3022.71	2019.27	1607.50	3152.88	3131.29	2668.30	2348.29	753.55
	G2[2]	1004.07	1317.86	2366.47	2586.49	2204.48	1178.57	2074.16	-	1818.87	636.11
Modulus of rupture (MPa) - MOR	G1[3]	106.52	69.57	139.11	109.25	84.67	203.12	138.99	104.18	119.43	41.35
	G2[4]	85.78	66.40	89.97	130.11	107.94	61.01	100.88	-	91.72	23.95

[1] Obtained using Equation 1; [2] Obtained using Equation 2; [3] and [2] obtained using Equation 3; BP: body of proof.

due to the lack of significant differences in their geometrical dimensions in spite of the difference in their transversal sectioning. Interestingly, groups G3 and G4 demonstrated similar results for resilience to flexion. This could be considered an advantage when using hollow rods, as the same resistance of a solid rod can be obtained with less material. Moreover, other materials could be used to fill the hollow space of these rods and potentially increase their resilience while maintaining the ability of being perforated for the insertion of the locking screws.

A comparison between the mechanical properties of flexion tests from groups of femurs without osteotomy (G1 and G2) and those with rods (G3 and G4) demonstrated that the main mechanical property to be improved was the modulus of elasticity of rods. This property could be significantly improved by using reinforcing materials such as glass and carbon fibres, manufactured in a similar way or by other polyamides synthesized by other processes, such as injection moulding. SPADETO JUNIOR et al. (2011) reported faults on the rods at the site of the screw closest to the fracture line and fissures in the bone. In the present study; conversely, the tests from G5 and G6 have shown that faults on the rods

occurred close to the osteotomy site and not where the screws were inserted.

During a flexion test the bone is simultaneously compressed on one side and tractioned on the other. Due to its anisotropy, when flexed the fault, it will always be present on the side subjected to the traction (SPADETO JUNIOR et al., 2010). In this study, crack nucleation (bone-rod) was observed on the area tractioned, as the bone is less resistant to traction than compression. Similarly, SPADETO JUNIOR et al. (2010), in a study (*in vivo*) that used polyamide for the stabilization of induced fractures in the femurs of calves, have reported that half of the inserted rods had a fracture on the interface containing the screws close to the osteotomy line. In the present study; however, the rupture was observed in the region of the osteotomy and not by the screws. These findings are thought to be due to the good drilling technique applied, the tight insertion of the rods and the coaptation of the fracture to the type of polyamide and its manufacturing; which minimized the fatigue on the screws and evenly distributed the stress throughout the system, resulting on the fracture of the rod at its weakest point: the osteotomy site.

Table 2 - Flexion test results (Modulus of elasticity and rupture) of G3 (two solid polyamide 12 rods) and G4 (two hollow polyamide 12 rods).

	---Modulus of elasticity (MPa) – MOE--			
	BP1	BP2	Mean	Standard deviation
G3 [1]	1260.78	1501.84	1381.31	170.45
G4 [2]	1349.45	1267.32	1308.39	58.07
G3 [3]	77.20	87.36	82.28	7.18
G4 [4]	79.36	77.91	78.64	1.02

[1] Obtained using Equation 1; [2] Obtained using Equation 2; [3] and [4] obtained using Equation 3; BP: body of proof.

No significant difference (P<0.04) was observed in the mechanical properties between the bone-implant systems using solid or hollow rods (Figure 1G and 1H). This could be due to the fact that there were no significant differences in their geometrical dimensions in spite of the difference in their transversal sectioning. Groups G5 and G6 (bone-implant) did not reach the mechanical resistance levels of G1 (control). However, there was a synergism between the use of polyamide 12 rods and the interlocking system with screws, as the bone-implant system resisted load levels greater than those by rods alone (G3 and G4). These observations, in conjunction with the possibilities of reinforcing polyamide 12, provide great scope for future research, which could analyse the use of reinforced polyamide 12 rods manufactured and implanted in a similar manner to the ones described in this study.

The modulus of elasticity and the modulus of rupture during flexion could not be calculated for G5 and G6 as the intricate geometry of the structure did not allow the determination or application of a mathematical model that could accurately represent the physical state of the loads, geometry, boundary conditions and behaviour of the materials. Thus, approximate models, in which the principles of the elasticity theory could be applied in an accessible and precise manner, should be used. According to LEE (2015), the most commonly used method for this is the Finite Element Method (FEM), which subdivides the domain into simpler parts to solve the problem. Thus, herein a suggestion for a future study with the use of specific software, such as the ANSYS mentioned by LEE (2015), to correctly determine the values of the mechanical properties of the elements of the bone-implant system and also of the stress throughout the system.

CONCLUSION

The polyamide 12 rods proved to be adequate for implantation following bone drilling, as well as for interlocking with screws, thus making its use in live animals promising. The resilience of hollow rods did not differ from that of solid ones; therefore, future studies should look into incorporating other materials to give even greater resilience to the bone-implant system. The bone-implant system withstood load levels greater than the rods alone and these findings, in conjunction with the possibility of reinforcing the polyamide with other materials, could generate interesting studies.

BIOETHICS AND BIOSSECURITY COMMITTEE APPROVAL

The study was approved and conducted under the guidance of the Ethics Committee in the Use of Animals of the Universidade de Franca (UNIFRAN), protocol number 026/12.

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