Design and characterization of a biomimetic composite inspired to human bone

F. LIBONATI, C. COLOMBO and L. VERGANI

Politecnico di Milano, Department of Mechanical Engineering, Via La Masa 1, 20159 Milano, Italy

ABSTRACT Many biological materials are generally considered composites, made of relatively weak constituents and with a hierarchical arrangement, resulting in outstanding mechanical properties, difficult to be reached in man-made materials. An example is human bone, whose hierarchical structure strongly affects its mechanical performance, toughness in particular, by activating different toughening mechanisms occurring at different length scales. At microscale, the principal toughening mechanism occurring in bone is crack deflection. Here, we study the structure of bone and we focus on the role of the micro-structure on its fracture behaviour, with the goal of mimicking it in a new composite. We select the main structural features, the osteons, which play a crucial role in leading to crack deflection, and we reproduce them in a synthetic composite. The paper describes the design, manufacturing and characterization of a newly designed composite, whose structure is inspired to the Haversian structure of cortical bone, and that of a classic laminate developed for comparative reasons. We conclude with a critical discussion on the results of the mechanical tests carried out on the new composite and on the comparative laminate, highlighting strengths and shortcomings of the new biomimetic material.

Keywords bio-inspired; biomimetics; bone; composite.

NOMENCLATURE		
CF	=	carbon fibres
GF	=	glass fibres
NCF	=	non-crimp fabric
UD	=	unidirectional

INTRODUCTION

Many biological materials are generally considered composites, made of relatively weak constituents. However, their hierarchical arrangement results in a great combination of mechanical properties (e.g. stiffness, strength, toughness and low density), difficult to be reached in man-made materials. This has recently raised the interest of researchers in the study of these materials, in particular on their structure–property relationship. Indeed, there is an increasing interest in the materials research community in understanding the mechanisms governing the behaviour of natural materials, with the aim of reproducing them in *de novo* synthetic materials.[1–3] This research area is called biomimetics[4] and has led to many breakthroughs in material science. It is an interdisciplinary field of research, which has recently found many engineering

applications. Biomimetics is a design approach to create new materials and solve technical issues, by mimicking structures existing in nature.[5,6] Indeed, nature is full of many smart systems, which can be easily copied, also with the aid of recent technologies. In the last decades, the biomimetic approach has led to a large variety of solutions, inspired to nature, also known as bio-inspired or biomimetic structures. Examples of biomimetic systems present in the literature are Velcro, a fastening device inspired to the hooking system of the burdock plant, patented in 1955,[7] self-cleaning super hydrophobic surfaces inspired to the lotus leaf,[8] woven fabrics for swimsuits, designed to reduce water drag, by mimicking the characteristic scales of shark skin,[9] nanocomposites reproducing the structure of nacre,[3,10] self-dry adhesives inspired to the hierarchical structure of the gecko foot.[11,12]

Nature generally uses hierarchical structures, to ensure adaptation to different functions.[6] Indeed, many natural systems are the result of a self-assembly process, which

Correspondence: F. Libonati. E-mail: flavia.libonati@polimi.it

leads to a hierarchical organization with multiscale dimensions of features, ranging from the molecular scale up to the macroscale. This multiscale hierarchical structure generally ensures adaptation of each organization level to specific functions.

Among biological composites, wood, bone and nacre, also known as biominerals, are considered interesting structural materials, for their load bearing capacity. Recently, these materials have also been considered biomimetic models, yielding to the development of new biomimetic solutions.[1] In particular, bone, which provides support for many organisms, is mostly attractive, for the optimal combination of mechanical properties (i.e. stiffness, strength and toughness). Indeed, bone, one of the most ancient and common biological materials, has been the object of research for many years and currently constitutes a source of inspiration for many biomimetic materials.[13] Nowadays, it is still considered a very intriguing material and currently studied by many research groups, either in the field of medicine, to understand its properties and behaviour for the development of proper treatments, or in the field of engineering, to design bio-inspired materials for tissue engineering and regenerative medicine,[14] but also for structural applications.[3,13]

The noticeable mechanical properties of bone, such as stiffness, strength and toughness in particular, make it an attractive biomimetic model for the development of *de novo* biologically inspired composites. Bone, which can be simply considered as a mineral–protein composite, is well known for its remarkable toughness. This mechanical property is about three to five orders of magnitude more than that of its constituent mineral and has its origin at different length scales, hence at different hierarchical levels, where particular toughening mechanisms generally occur.[15]

Because no man-made composite has gained such an increase in toughness, with respect to the basic constituents,[16] it is interesting to see whether we can have a new composite material with improved toughness, by mimicking the structure of bone. The aim of this study is to reproduce some of the toughening mechanisms occurring in the microstructure of human bone, by mimicking the microstructural design in a *de novo* composite material.

In the following, we present the design, realization and testing of a new composite material, inspired to the microstructure of human bone, by using a biomimetic approach as schematically represented in Fig. 1. The bone-inspired composite is a synthetic material made of glass and carbon fibres (CF) embedded into an epoxy matrix and showing an internal organization aimed at reproducing the main structural features occurring at bone microlevel (i.e. osteons). We show the results of a complete characterization performed on this material and on a classic laminate, made of the same type and amount of fibres and resin, used for comparative reasons. After testing both the materials, we finally propose new solutions to improve the initial design, based on the results of the experimental tests and on numerical simulations previously performed on bone nanocomposites.

BONE AS A BIOMIMETIC MODEL

Bone is a structural biological material, which provides structure and support for many organisms. It is a hierarchical composite, made of two main constituents, collagen matrix and hydroxyapatite mineral crystals, the former providing elasticity and the ability of dissipating energy under mechanical deformation and the latter providing load bearing capacity. Collagen protein and hydroxyapatite crystals are considered the basic building blocks and are present in all bony tissues.[17]

Bone, although its rather weak constituents, has noticeable mechanical performance, mainly determined by its structure. Indeed, the internal structure of bone is quite complex and characterized by a hierarchical organization, where we can distinguish seven levels of hierarchy, ranging from the atomistic to the macroscale.[18] Each level is characterized by particular features, whose size and arrangement have a key role in determining the properties of the whole material.

At sub-nanoscale, we can recognize the above-mentioned building blocks: the hydroxyapatite crystals, hexagonal lattice crystals mainly made of calcium and phosphate, and the tropocollagen protein, coiled into a
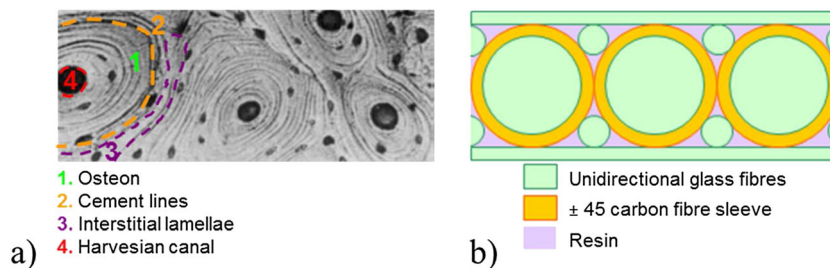

Fig. 1 (a) Bone microstructure and (b) internal structure of the bio-inspired composite.

triple helix. Besides, we can also recognize other non-collagenous proteins. At nanoscale, tropocollagen molecules are assembled together with non-collagenous protein in a hydrated environment, to form collagen fibrils, placed in a staggered configuration. This configuration is characterized by gap regions, which are filled, during the mineralization process, by hydroxyapatite crystals, to form mineralized collagen fibrils. At the micrometer length scale (~10 μm), the mineralized fibrils are organized in fibril arrays, held together by a protein phase and arranged in different patterns, for example, parallel, random and woven, to form lamellae. Concentric lamellae form, at a larger level, cylindrical structures (with a diameter up to 100–200 μm), called osteons, which are the characteristic features of the bone microstructure, also known as secondary or Haversian structure. The osteons present an internal canal, called Haversian canal (50–90 μm), responsible for the primary bloody flow and small transversal canaliculi, providing the secondary bloody flow. The osteon outer layer, called cement line, is about 1–5 μm thick and results from remodelling process. In fact, bone Haversian structure is also known as secondary structure, as it results from remodelling, whereas bone primary microstructure generally originates during growth and is characterized by smaller and shorter osteons without cement lines. Besides concentric lamellae, at this level it is also possible to find interstitial lamellae, placed in between different osteons.

At macroscale, bone structure starts to differentiate in cortical, or compact bone, and trabecular, or spongy bone, differing for the density and consequently, for their mechanical properties. This differentiation of the bone tissue is a typical example of adaptation to different functions.

The mechanical behaviour of bone varies with the hierarchical scale. Thus, we can distinguish the tissue-level mechanical behaviour, from the global macroscale behaviour. Moreover, at each level, we can notice particular mechanisms, which define the tissue-level performance and affect the overall behaviour of bone. For instance, at microstructural level, we can find the principal toughening mechanisms, affecting the fracture behaviour of bone: the so-called *extrinsic toughening mechanisms*, largely studied in the literature.[18,19] These mechanisms include microstructural processes that inhibit the crack growth process, by increasing the dissipation ability of bone, thus the fracture toughness. Nevertheless, according to Launey *et al.*,[19] bone toughness results from a reciprocal competition of toughening mechanisms occurring at the microscale. In the range of 10–100 μm, which corresponds to the osteon level, the main toughening effects result from crack deflection and twisting, crack bridging by uncracked ligaments, constrained microcracking and collagen-fibril bridging. Nalla *et al.*[20] state that these mechanisms, mainly affecting the crack growth, have a different contribution in increasing the toughness. However, the largest contribution to the increase in toughness is given by crack deflection.[21] A key role in crack deflection is thought to be played by the osteons, which deflect and twist the crack along their outer boundaries, also known as cement lines, which are also sites of microcracks nucleation.[22] Indeed a crack, while propagating orthogonally to the main osteon direction, generally changes its direction once it reaches a cement line. The change in the crack path results in an increase in the energy required for crack propagation, hence an increase in toughness.

BIO-INSPIRED DESIGN: MATERIALS AND MANUFACTURING

In this section, we describe the biomimetic approach used to mimic the microstructure of bone in a *de novo* composite material, from the initial concept to the final design and manufacturing. The approach followed to mimic the bone internal structure consists of different phases: (i) the choice of the hierarchical level to be reproduced, in order to mimic some particular toughening mechanisms; (ii) the definition of a concept; (iii) the feasibility assessment; (iv) the choice of materials and manufacturing technique; and (v) the realization. In particular, the phases (ii), (iii) and (iv) are strictly connected, because the initial design was simplified to make it feasible with respect to the available manufacturing techniques. In phase (iv) during materials selection, we also consider the costs of materials, with the aim of proposing a cost-effective solution, compared with other conventional composites used for structural applications. Besides the bio-inspired composite, we also show the design and characterization of a classic laminate, characterized by the same type and amount of reinforcement and matrix of the bio-inspired one, but having a different internal organization (i.e. conventional lay-up). The laminate was produced and tested with the aim of allowing a complete comparison with the bio-inspired material.

The hierarchical level, chosen as a model for the bio-inspired material is the microstructural level, which is characterized by the presence of a repeating unit, called osteon, with a cylindrical shape. Each osteon is made of concentric lamellae with an internal vascular canal ensuring the metabolic activity. The microstructure of bone has been largely studied in the literature, by experimental measurements under different loading conditions (i.e. tension, compression, bending and torsion).[23–26] These studies confirmed a marked anisotropy characterizing

the mechanical properties of bone, mainly due to the longitudinal orientation of the majority of fibres in the osteons.[27]

The design of the bio-inspired composite material is not proposed as a mere copy of the microstructure of bone. It is a simplified structure, which contains the main structural features involved in the fracture process, the osteons, reproduced at larger scale, by means of synthetic fibres. By mimicking bone structure, we aim to replicate the main toughening mechanisms occurring in bone at this hierarchical level (i.e. crack deflection and longitudinal splitting). Our goal is to find a method to improve the toughness of synthetic composites materials, which are known to be quite brittle materials. The proposed design is given in Fig. 1, along with a picture showing the microstructure of bone. The main structural features, replicated in the biomimetic structure are colour highlighted.

The design includes many simplifications: first, being the bio-inspired material made of synthetic constituents, living functions, which are responsible of bone remodelling and self-healing capacity, cannot be provided by that (e.g. we do not replicate the Haversian canal, responsible of the primary bloody flow); besides, we introduce other simplifications to be able to reproduce the design with the available manufacturing tools.

Materials

In our design, we chose to replicate the main structural features, which are thought to play a crucial role in the fracture behaviour. We replicated osteons with unidirectional (UD) bundles of glass fibres (GF; Roving 2400 tex by 3B fibreglass company, Battice, Belgium), offering resistance to axial loadings. This is a large simplification, because the geometry of the continuous fibres is completely different from that of concentric lamellae; nevertheless, the simplified system aims to mimic the function of the concentric lamellae. External lamellae, called cement lines, were implemented in the composite, by means of sleeves made of ±45° CF (Torayaca® T300 1 k by Toray Carbon Fibers America, Santa Ana, CA, USA), which collect and pack each osteon, preserving the fibre alignment. Besides offering bending strength, these tubular layers aim to reproduce one of the main properties of bone: the possibility of deflecting and twisting cracks. We reproduced interstitial lamellae by means of longitudinal UD-GF impregnated into an epoxy matrix, offering compactness and filling up the gaps between osteons. Then, we used two external layers of UD-GF-non-crimp fabric (NCF) (92145 Finish FK 144 by P-D Interglas, Erbach, Germany) to reproduce the outer circumferential system (i.e. circumferential lamellae), which packs the osteon architecture in human cortical bone. The two skins of UD-GF-NCF were placed at the top and the bottom of the composite structure, with

the aim of packing the internal tubular structure and offering a final flat and uniform surface for the composite plate. Here, we further simplified our design, replacing the circumferential structure with a flat one, both for manufacturing reasons and also to allow a larger variety of applications of these composite panels in the structural field. Another significant approximation is the scale difference (about one order of magnitude) between the osteon diameter size (100–200 μm) and the sleeves diameter size (4–5 mm).

In addition to the bio-inspired material, we also produced another laminate, to allow a direct comparison with the newly designed material. The comparative material is made of the same constituents of the bio-inspired composite (UD-GF, ±45° CF and epoxy resin) in the same ratios. In both materials, the fibres have the same orientations, and the UD fibres are placed in the outer layers. The constituents of the two analysed composites are the same, so are the properties. The difference lays in the internal arrangement: the biomimetic composite is characterized by a repeating cylindrical unit, which aims to mimic the osteon, whereas the laminate is characterized by the overlap of different layers, resulting in the following stacking sequence: $[GF(0°)_4, CF(\pm45°)_2, GF(0°), CF(\pm45°)]_s$. The GF outer layers and the CF layers are both NCF, whereas the internal layer of GF are UD fibres. Both the materials are characterized by 54%vol. fibres, consisting of about 6.5%vol. CF and 47.5%vol. GF. We should stress that the comparative material is not optimized from the strength point of view because it is not designed for a specific loading direction/application.

Manufacturing

The manufacturing process has been developed in cooperation with Clausthal University of Technology (Germany). Due to the characteristic osteonic structure to be replicated by the bio-inspired composite, a particular procedure and equipment were developed and used in the manufacturing stage. Many trials were performed to assess proper equipment for the panel's preparation. Indeed, it appeared immediately evident, from the first stages of production, that most of the required time was necessary for osteon preparation and alignment. A special frame made of a wooden plate and nails was used, for fibres disposition with fixed length. During the fibres preparation phases, before resin injection, it was essential to preserve fibre alignment, and the use of such a tool was crucial. Details of fibres placement are shown in the images of Fig. 2. As shown in these pictures, each osteonic cable, made of tubular woven CF (diameter 5 mm and thickness 0.18 mm), is previously filled up in UD-GF. Among these operations, most of the time was spent in the final manual stitching.
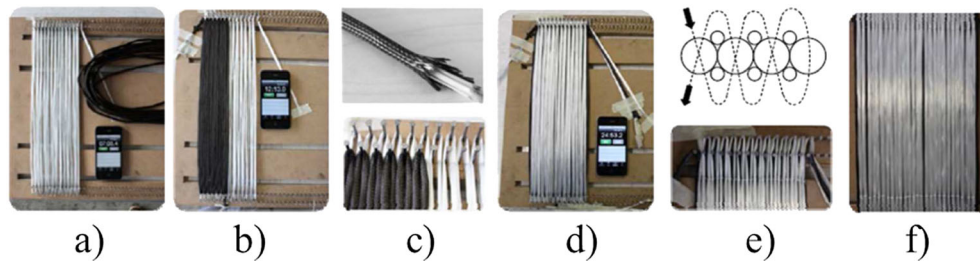
Fig. 2 Details of the bio-inspired material manufacturing process: a) first glass roving non-crimp fabric disposition, stretch, fixing; b) osteon cable overlap; c) detail of the fibre fixing and the carbon sleeve filling; d) last glass roving overlap; e) manual stitching; and f) sew, fixing and ends cutting.

Also for resin injection, a new system was designed, exploiting features from vacuum assisted resin injection, as the use of a pump, and from resin transfer moulding (RTM), as the use of a mould. Advantages of this developed process are easiness of centring and prevention of runners; moreover, it avoids bending and depressurization problems and ensures flatness to the final product.

Dimensions of manufactured plates are 110×50 mm; a view of the cross-section of one of these plates is given in Fig. 3, where osteons are sufficiently regular. However, the presence of many defects in the end products must be considered, being this new manufacturing technique completely manual.

In parallel to the osteonic plates, also a classic laminate was manufactured. The manufacturing process included different phases: the placement of UD-GF by filament winding, the placement of CF-NCF and the placement of UD-GF-NCF at the top and the bottom to ensure flat surfaces.

CHARACTERIZATION

This section describes the experimental mechanical characterization of the two materials, the bio-inspired synthetic composite and the classical laminate. Different kinds of tests were selected in order to perform a full characterization of mechanical properties in two main directions, especially for the osteonic structure: that along osteons and the perpendicular one. The aim of these tests was to compare mechanical properties of the two materials and to stress the attention on strength and shortcomings of the new designed material.

Fig. 3 View of the cross-section of the manufactured bio-inspired composite structure.

Mechanical testing

We performed a complete characterization of both the materials in longitudinal and transversal directions. Longitudinal direction is considered as the one along UD fibres.

Indeed, we carry out tests under different types of static loading conditions:

1 tensile tests, according to American Society for Testing and Materials (ASTM) D3039/D3039M-08,[28] with a cross-head speed of 2 mm/min and a data acquisition frequency of 5 Hz; for tensile tests in longitudinal direction, we used a Schenck servo-hydraulic machine, endowed with a 250 kN load cell, whereas for tensile tests in transversal direction, we used an MTS Landmark servo-hydraulic machine (Eden Prairie, MN, USA), endowed with a 100 kN load cell;

2 compressive tests, according to ASTM D3410/D3410 M-03,[29] using a cross-head speed of 1.5 mm/min and 5 Hz as data acquisition frequency; for these tests, we used the same machine adopted for the longitudinal tensile tests;

3 flexural bending tests (i.e. three-point bending configuration), according to Ente Nazionale Italiano di Unificazione (UNI) European Committee for Standardization (EN) International Organization for Standardization (ISO) 14125,[30] adopting a cross-head speed of 2 mm/min and 5 Hz as data acquisition frequency; for these tests, we used an MTS Alliance RT/100 machine endowed with a 25 kN load cell; also, a deflectometer (MTS model 632-06H-30) was used to measure the mid-span deflection;

4 fracture toughness tests, according to ASTM E1922-04,[31] using a cross-head speed of 1 mm/min; the load is applied through pin-loading clevises and the displacement at the notch mouth measured by using a displacement gage attached to the specimen through knife edges. For these tests, we used an MTS Alliance Rf-150 machine endowed with a 90 kN load cell.

In cases 1–3, we adopted rectangular specimens, with adhesively bonded tabs for the first two cases; for case 4 instead, we adopted single-edge-notch specimens (length = 100 mm; width = 25 mm; thickness = 5 mm) in

opening mode loading. To create a narrow notch, which extended through the thickness and over half of the specimen width, we used a diamond-impregnated copper-slitting saw.

Beyond these tests, we also carried out interrupted fracture toughness tests, by using the same type of specimens adopted for case 4. We monotonically applied the load, and interrupted at each 0.5 kN step for few minutes, to allow microscopic analyses to be performed by means of an optical microscope and image acquisition with the aid of a laptop.

Microscopic analyses

We performed microscopic analyses during the interrupted fracture toughness tests, by using an optical microscope endowed with LEICA DFR 290 lens (Wetzlar, Germany) and a computer with a suitable software for image acquisition.

We also analysed the cross-sections of the specimens subjected to fracture toughness tests, by means of a scanning electron microscopy (SEM). We cut the specimens in the vicinity of the zone, where the crack had previously propagated, and we observed the cross sections with an SEM Evo 50 EP Zeiss by Oxford Instrument (Carl Zeiss SMT, Germany). Before microscopic analyses, we

cleaned the sample surfaces and made them electronically conductive, by sputter-coating with a conductive material (i.e. gold).

RESULTS

Mechanical testing

Characteristic results of each mechanical test are shown in the plots of Figs 4–6 and 8, presenting stress–strain, stress-crosshead displacement and force-notch mouth displacement curves.

Figure 4a represents the experimental stress–strain curve of two characteristic specimens from the two materials: It is related to the tensile test along longitudinal direction. It appears evident that the bio-inspired composite experiences higher performances, both in terms of ultimate strength and stiffness, which corresponds to the slope of the two curves. The bio-inspired composite, stiffer, fails in correspondence of a lower maximum strain, with respect to the classical laminate. The organization of synthetic fibres in the composite, inspired to the bone structure, allows therefore to have higher performances, but lower deformation capability.
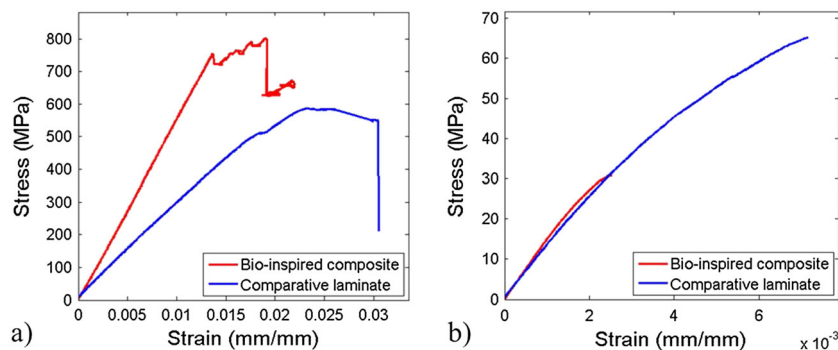

Fig. 4 Stress–strain curves during tensile static tests on bio-inspired and comparative composite: a) longitudinal direction and b) transversal direction.
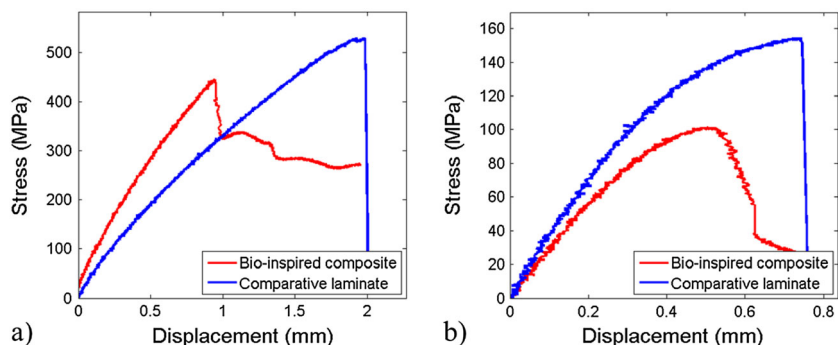

Fig. 5 Stress–strain curves during compression static tests on bio-inspired and comparative composite: a) longitudinal direction and b) transversal direction.
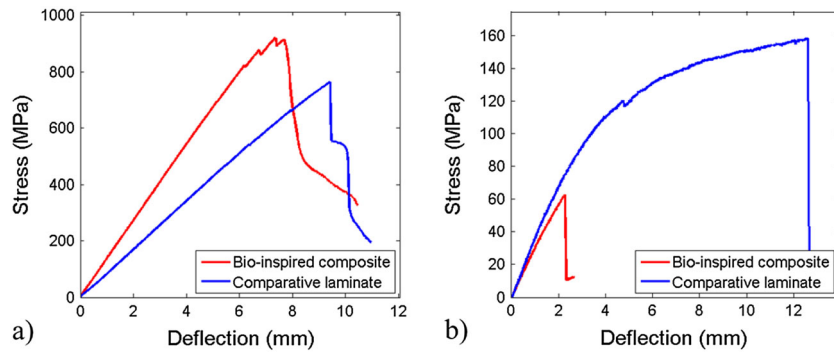
Fig. 6 Stress-deflection curves for longitudinal and transversal directions during three-point bending static tests on bio-inspired and comparative composite: a) longitudinal direction and b) transversal direction.

The osteonic structure, instead, is detrimental for its mechanical properties along transversal direction. Figure 4b shows the stress–strain plot of the static tensile test along the transversal direction. The mechanical behaviour of the osteonic structure is weaker than the classic laminate. Indeed, even if the stiffness is comparable, the final failure of the bio-inspired composite occurs at a low stress value, less than half the laminate ones. The interplay of the various laminate layers, indeed, seems to enhance the transversal properties. In the bio-mimetic structure, cracks propagate between the cylindrical osteons, mainly at the interface between CF tubular layers (outer part of the osteon) or in the resin. In this region, it seems that the resistance to crack propagation is the lowest.

Similar considerations can be drawn for the static tests performed in compression and in bending, whose results are presented in Figs 5a and b and 6a and b, respectively.

For compression, the osteonic composite is still stiffer in longitudinal direction, but the ultimate strength is lower in absolute value than that of the comparative structure (Fig. 5a). In transversal direction (Fig. 5b), the osteonic structure experiences a slightly lower stiffness, but the question of crack propagation at the osteon interface remains a drawback, resulting in a deeply lower ultimate strength.

Tests in bending configuration (Fig. 6a & b) show very similar results to tensile tests. Plots are reported in terms of stress deflection. Comparing flexural to tensile strength, the osteonic structure has similar performances in longitudinal direction, but doubled along the transversal direction. The laminated composite, instead, improves its strength both in longitudinal and in transversal directions.

Also, failure modes are similar to tensile tests: Examples along transversal direction are shown in Fig. 7. The two considered materials experience completely different failure modes, dictated by their inner structure, and cracks propagate in the osteonic or in the laminate structure along different paths, thus requiring different failure energies. Compared with the classic laminate,
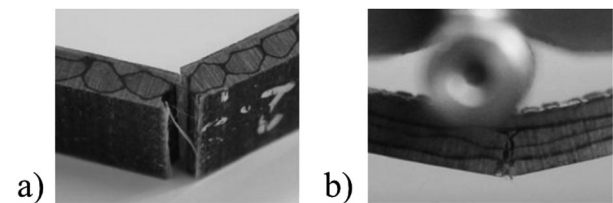

Fig. 7 Flexural bending tests in transversal direction: a) failure mode of the bio-inspired composite and b) failure mode of the composite laminate.

the bio-inspired composite shows a different macroscopic mechanical behaviour, related to the failure mode, and owing to the different internal organization. The osteonic structure shows some shortcomings in transversal direction, but it maintains higher transversal tensile stiffness if compared with the conventional material.

Table 1 shows a summary of the results from experimental tests. It should be mentioned that all performed tests show a good repeatability of the results. For each type of test, five specimens were used, except for fracture toughness tests, where we used only three specimens. This table also reports the results of the fracture toughness tests; a typical plot obtained from experimental tests is shown in Fig. 8 for the two considered laminates. From these experimental tests, it is possible to state that the laminate composite has a slightly better response to fracture in presence of a crack. Fracture toughness and fracture strength are respectively 18% and 16% higher than the bio-inspired composite. It is interesting to see how the presence of a crack affects the strength of the bio-inspired composite, by reducing it to less than a half. In the case of conventional composite, instead, the strength of the material in presence of a crack is 20% lower than the correspondent flawless material. The lower value of the fracture strength in the bio-inspired material is due to reinforcement, which is mainly longitudinal. Also, being the ±45° CF in a tubular shape, they did not

Table 1 Results of the experimental tests performed on the bio-inspired composite and on the comparative laminate in both longitudinal and transversal directions

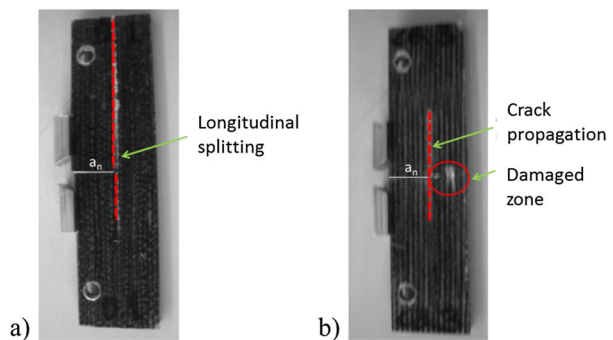| Property	Unit	Longitudinal		Transversal	
		Bio-inspired	Comparative	Bio-inspired	Comparative
Tensile strength	MPa	797 ± 53	568 ± 10	28 ± 3	65
Compressive strength	MPa	416 ± 32	581 ± 48	101 ± 2	157 ± 2
Flexural strength	MPa	880 ± 3	782 ± 3	59 ± 5	156 ± 2
Tensile modulus	MPa	$46\,486 \pm 4981$	33 245	$14\,558 \pm 972$	12 366
Compressive modulus		↑	↓	↓	↑
Flexural modulus	MPa	$44\,296 \pm 2195$	$31\,108 \pm 595$	$10\,585 \pm 2061$	$14\,583 \pm 104$
Fracture toughness	MPa$\sqrt{m}$	26.87 ± 2.93	32.68 ± 1.12	—	—
Fracture strength	MPa	379 ± 43	452 ± 17	—	—
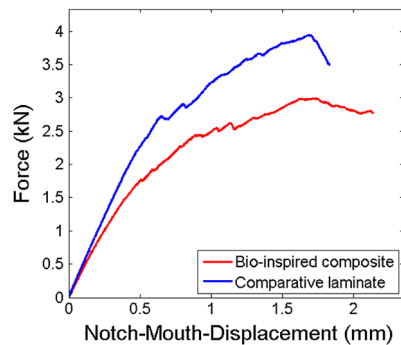
Fig. 8 Comparison between the fracture behaviour of the bio-inspired composite and that of the comparative laminate.

Fig. 9 Comparison between the fracture behaviour of the bio-inspired composite and that of the comparative material: the crack path is highlighted with a red dashed line. a) Failure of the bio-inspired composite: crack splitting and b) failure of the comparative composite: crack propagation and damage area ahead the crack tip.

contribute to prevent the crack growing, as the ±45°-CF textiles (i.e. NCF) did for the comparative laminate.

In the bio-inspired composite an inter-osteon reinforcement, which keeps the sleeves together allowing them to work as a unique system and preventing the crack growth, is missing. The initial design behaves as a discrete system, where a lack of continuity occurs between the osteons. Despite these drawbacks, the bio-inspired material has shown to replicate the typical toughening mechanisms occurring in the microstructure of bone (i.e. crack deviation and longitudinal splitting). Figure 9 shows a comparison between the failure mode of the bio-inspired material and that of the comparative one, after a fracture toughness test. In the comparative material, failure occurred because of fibre–matrix debonding and delamination; whereas in the bio-inspired material, the crack seemed to propagate at the interface between two osteons, leading to a crack splitting phenomenon. This hypothesis is also confirmed by the results obtained from microscopic analyses presented in the following paragraph.

Microscopic analyses

The bio-inspired material has shown to reproduce some typical toughening mechanisms occurring in

the microstructure of bone (i.e. crack deviation and longitudinal splitting). This mechanism is clear from the interrupted fracture toughness tests, which allow the fracture process to be followed. During the tests, we observed the formation of many small cracks, whereas larger cracks showed deviation and leading to splitting and failure.

To better understand the effect of the osteon structure, we also created notches with different initial length and with the crack tip ending in both the inter-osteon and the intra-osteon regions. In the former case, the crack directly propagated along the osteon–osteon interface, whereas in the latter, the crack propagated in the osteon region, then, it deviates along the osteon–osteon interface, leading to longitudinal splitting.

These hypotheses, made on the basis of the observations performed with the optical microscope, are also confirmed by the SEM analyses. The results of the SEM analyses are shown in Fig. 10; the images are referred to the cross-section of a sample of bio-inspired composite and show the region where the main crack propagates. In this figure, the phenomenon of crack
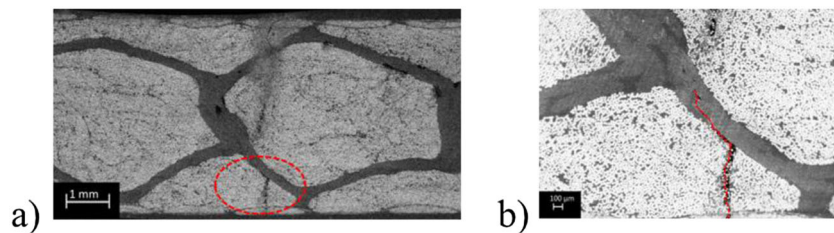
Fig. 10 Scanning electron microscopy images from backscattered electrons showing a cross-section of a sample of bio-inspired material. a) The image (magnitude 40×) shows the region where the main crack propagates; the crack region is highlighted with a red dashed line circle. b) The image (magnitude 100×) shows the crack deviation, from the intra-osteon to the inter-osteon region; the crack path is highlighted with a red dashed line circle.

deviation is clearly visible. The crack initially propagated inside the osteon, then, once it reaches the "cement line", it deviated. This mechanism is close to what occurs in human bone.[18–20]

CONCLUDING REMARKS

In this paper, we showed the design and realization of a new bio-inspired composite material, by means of a biomimetic approach. The material aims to reproduce some of the toughening mechanisms occurring in human bone at microscale, by mimicking the microstructure. In our work, we simplified the structure, selecting the main structural features involved in the fracture process and neglecting the living part of the bone tissue, and we used synthetic materials, such as GF and CF and epoxy resin to realize the material. Besides the bio-inspired material, we also showed the realization of a conventional laminate, made with the same type and amount of fibres and resin, and we tested both the materials, under different loading conditions to have a comparison.

The new designed biomimetic composite showed a different mechanical behaviour with respect to the conventional one, owing to the different internal structure. As expected, the bio-inspired structure is characterized by a marked anisotropy, with improved properties (i.e. stiffness and strength) in the longitudinal direction (parallel to the osteon main axis), except for the compression strength. In the transversal direction, it shows some drawbacks, although maintaining a higher tensile stiffness.

From the microscopic analyses, performed with an optical microscope and with an SEM, we could observe the occurrence of crack deviation, leading to a final splitting in the bio-inspired material, similarly to what generally occurs in human bone. Hence, despite its shortcomings, mainly due to the lack of continuity or reinforcement in the inter-osteon region, the newly designed material is able to mimic the typical bone fracture mechanism.

As future perspective, we aim to introduce further improvement in the initial proposed design, to limit the weaknesses in the transversal direction response and to improve the fracture strength, leading to an optimal biomimetic solution with high performance. In particular, we aim to improve the transversal behaviour, by enhancing the osteon–osteon interactions, for instance by creating a multilayer osteon structure, allowing simultaneous inter-osteon interactions in different directions. Another possible solution can be given by the addition of a weave composite fabric, alternatively placed under and over each osteon. The proposed design could be further optimized by adding reinforcing nanoparticles, with a platelet shape and a proper characteristic sizes, as suggested by the results of molecular dynamics simulations. Indeed, a previous study[32], Libonati *et al.* have shown that flaw tolerance occurs in hydroxyapatite nanocrystals below a certain size, acting as toughening mechanism at nanoscale. Thus, by adding platelet-like nanoparticles, we could reproduce the stiffening and toughening effects given by hydroxyapatite nanocrystals in bone at nanoscale. Moreover, a "glue-like system", providing "sacrificial local failure", can also be introduced in the new design to improve interface strength, mimicking the role played by hydrogen bonds in bone nanocomposites, as resulted from molecular dynamics simulations shown in a previous work.[33] Thus, we propose further improvements focused on two main aspects: the enhancement of interface strength, by using a "glue-like" system and the inclusion of platelet-like nanoparticles with a high aspect ratio.

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