Single flat-elongated and helical lamellar crystals have been grown thermotropically in a main-chain non-racemic chiral liquid crystalline polymer which was synthesized from (R)-(−)-4′-(Omiga-[2-(p-hydroxy-o-nitrophenyloxy)-1-propyloxy]-1-nonyloxy)-4-biphenyl carboxylic acid, PET(R*)-9. The crystals possess the identical orthorhombic lattice dimensions of a = 1.07 nm, b = 0.48 nm, and c = 5.96 nm determined by both electron diffraction (ED) in transmission electron microscopy (TEM) and wide angle X-ray fiber diffraction (WAXD) methods. Dark field (DF) image, bright field image, and selective area ED (SAED) experiments using TEM provide chain orientation information in both of these crystals. In the flat-elongated lamellar crystals, the chain direction is perpendicular to the substrate surface in a center zone along the long (b-) axis of the crystals. Moving away from this zone along the short (a-) axis of the crystal, the chain direction continuously tilts in the ac-plane. A small tilt of approximately 0.002° per molecular layer is estimated using the SAED results. In the helical lamellar crystals, the main twist direction is parallel to the helical axis, and the rotation angle for each molecular layer is approximately 0.05°. However, specifically designed DF experiments using the entire and partial (205) and (206) diffraction arcs show that the chain orientation direction is also twisted along the short helical axis of the lamellar crystal. The rotation angle is approximately 0.01° per molecular layer. Therefore a second twist direction with a changing molecular orientation exists in addition to the long helical axis of the crystal. Based on these experimental observations, the concept of a double-twisted molecular orientation in the helical lamellar crystal can be established, although in principle, the macroscopic translational symmetry is broken along both of the long and short axes of the helical lamellar crystals in Euclidean space. It has also been found that different numbers of methylene units in the backbones can substantially affect the chiral crystal structures and morphologies.