Supplementary material for: Rusmirović, J.D., Ivanović, J.Z., Pavlović, V.B., Rakić, V.M., Rančić, M.P., Djokić, V., Marinković, A.D., 2017. Novel modified nanocellulose applicable as reinforcement in high-performance nanocomposites. Carbohydrate Polymers 164, 64–74.
https://doi.org/10.1016/j.carbpol.2017.01.086

Novel modified nanocellulose applicable as reinforcement in high-performance nanocomposites

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2.1. Materials

Cotton used for NC isolation was supplied from AD. Niva, Serbia (Turkish origin). The following chemicals were supplied from Sigma Algrich: H_2SO_4 (PubChem CID:1118), ethanol (PubChem CID:702), methanol (PubChem CID:887), glacial acetic acid (GAA) (PubChem CID:176), toluene (PubChem CID:1140), HCl (PubChem CID:313), perchloric acid (PubChem CID:24247), maleic anhydride (MA) (PubChem CID:7923), oleic acid (OA) (PubChem CID:445639), tetrahydrofurane (THF) (PubChem CID:8028), ethylenediamine (EDA) (PubChem CID:3301), potassium hydroxide (KOH) (PubChem CID:14797), diethyl ether (PubChem CID:3283), sodium sulfate (PubChem CID:24436), pyridine (PubChem CID:1049), p-toluenesulfonyl chloride (PubChem CID:7397), dicyclohexylcarbodiimide (DCC) (PubChem CID:10868), dichloromethane (DCM) (PubChem CID:6344), 4-dimethylaminopyridine (DMAP) (PubChem CID:14284), styrene (PubChem CID:7501), 2-butanone peroxide (methyl ethyl ketone peroxide; MEKP) (PubChem CID:3672772), cobalt octoate (Co-oct) (PubChem CID:8696) and potassium bromide (KBr) (PubChem CID:253877). Commercial carbon dioxide (99.99% purity) (PubChem CID:280), nitrogen (99.99% purity) (PubChem CID:947) and helium (99.99% purity) (PubChem CID:23987) were supplied by Messer-Tehnogas (Serbia). Propylene glycol (1,2-propanediol) (PubChem CID:1030), xylene (mixture of 1,2-, 1,3- and 1,4-dimethyl-benzene) (PubChem CID:7929) and tetrabutyl titanate (TBT) (PubChem CID:21801) were supplied by Fluka. All the chemicals used in this study were of analytical grade and used as received. Waste poly(ethylene terephthalate) (PET), used for unsaturated polyester resin production, was collected from soft beverage bottles. PET bottles were crushed into small pieces (app. 0.5 × 0.5 cm) and then washed with ethanol and DCM to remove impurities and residual adhesives.

2.1.1 Nanocellulose (NC) isolation

NC was obtained from cotton by acid hydrolysis with 64 % H_2SO_4. In an Erlenmeyer flask of 1000 ml, 20 g of cotton and 200 ml of H_2SO_4 solution of different concentrations were added dropwise in an ice bath and stirred. The suspension was heated while stirred if necessary to reach the appropriate temperature (40 min, 40 ^oC). Afterwards, the cotton dispersion was washed with deionized water using repeated centrifugation (n=6000 rpm/min) and sonication cycles, *i. e.* the supernatant was removed from the precipitate and replaced by fresh deionized water, and cycle was repeated. The centrifugation step was repeated until pH value of 4 is achieved or the supernatant became turbid. The last wash was conducted using dialysis with deionized water until the wash water maintained at constant pH or washed with deionized water until the pH value reached 5.5. The NC isolation procedure is shown on **Fig. S1.**

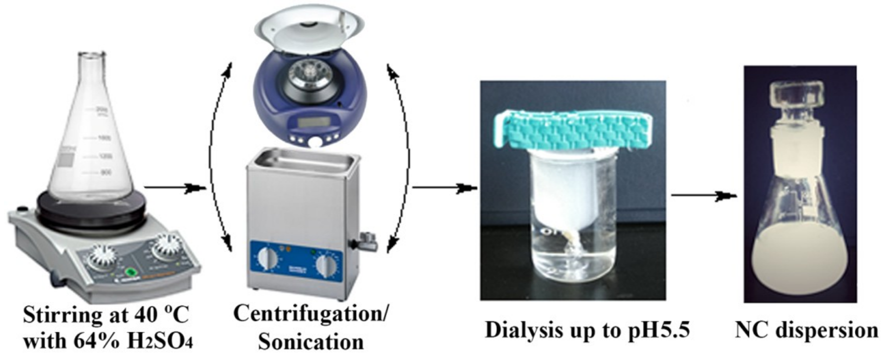

Fig. S1. Preparation of NC

2.2 Isolation of fatty acid of linseed oil (LO) and sunflower oil (SO)

In a four-necked glass reactor of 2 l, equipped with a reflux condenser, mechanical stirrer, thermometer and dropping funnel, 233 g (0.26 mol, 250 ml) of linseed oil (LO) dissolved in 700 ml of 96% ethanol was added. The KOH solution in ethanol (30%, 0.91 mol) was slowly added to oil with intensive mixing, and the obtained molar ratio of KOH/LO was 3.5/1. During the addition of KOH solution, the reaction mixture was maintained at constant temperature below 10 °C for an hour. Next two hours, the reaction mixture was heated to 50 °C, whereupon two-thirds of the solvent were removed by distillation. The resulting pasty residue was dissolved in the required amount of distilled water to get clear solution, and after addition of activated carbon and filtration, the obtained solution was acidified by addition of appropriate volume of 10% HCl until pH of 3 was attained. Top layer of FALO was separated; the aqueous layer was extracted twice with 250 ml of diethyl ether and combined with organic layer. The resulting ethereal solution was dried with sodium sulfate. Evaporation of ether by atmospheric distillation was followed by assembling vacuum system which enabled drying and removal of low boiling component of reaction product. The isolation of sunflower oil (SO) fatty acid, named FASO, was performed in an analogous manner.

2.3 Isolation of methyl ester of linseed, sunflower and soybean oil fatty acid

For isolation of methyl ester of LO, previously described in detail (Rusmirović et al., 2015), 929.0 g (3.3 mol) of LO were used, dissolved in 85 ml of methanol. The KOH solution in methanol (0.12 mol of KOH in 102 ml of methanol) was added dropwise. Afterward, the reaction mixture was heated at 58 – 62 °C for 3 hours, and then left to cool down. Bottom layer, *i.e.* mainly raw glycerin, was separated, and upper layer was treated with active charcoal and filtered through diatomaceous earth. After drying with sodium sulfate, the obtained linseed oil methyl ester was purified by vacuum distillation under nitrogen. The isolation of methyl ester of SO and soybean oil (SOYA) fatty acid fatty acid was performed in an analogous manner. Characteristics of fatty acids mixture (FA) and methyl ester of fatty acids (MEFA): acid value (AV), iodine value (IV), ester content and color are shown in **Table S1**.

Table S1
Characteristics FA and MEFA of LO, SO and SOYA

Fatty acids	*AV*, *mg KOH/g*	*IV*	Ester content, *%*	Color
FALO	200	152	-	Light yellow
FASO	189	147	-	Dark yellow to orange
MEFA/LO	5	152	97	Light yellow
MEFA/SO	4	144	94	Light yellow
MEFA/SOYA	7	131	96	Dark yellow to orange

2.5 Three step chemical modification of nanocellulose with MEFA *via* cross-linker

In order to change solvent for the modification, the obtained cellulose nanoparticles were washed with GAA, and 2 g of purified NC was charged into an Erlenmeyer flask of 250 ml. The 20 ml of GAA and 25 ml of toluene were added to purified nanocellulose and mixture was homogenized on ultrasonic bath (Bandelin electronic, Berlin, Germany, power 120 W, frequency 35 kHz) for 10 minutes. After homogenization, 0.1 ml of 60 wt.% perchloric acid and 0.5 g of MA were added into the mixture and left in ultrasonic bath for one hour. After completion of the reaction, the MA modified NC (NC-MA) was centrifuged and washed three times with toluene.

In the second step, NC-MA was modified with EDA. The NC-MA was placed in 250 ml four-necked flask and dispersed in 50 ml DCM for 10 min by using ultrasonic bath. After a condenser, thermometer, gas inlet tube and dropping funnel was assembled. In the NC-MA dispersion, mixed on a magnetic stirrer, was added 5.74 g of DCC and 0.73 g of DMAP in 30 ml DCM from dropping funnel providing inert atmosphere in a reaction system. Thereafter, dropwise addition of 1.2 g of EDA dissolved in 10 ml of DCM last for 30 min at room temperature, and continued with a mixing for 12 hours. After the reaction completion, modified product (NC-MA-EDA) was washed three times with DCM, and three times with toluene.

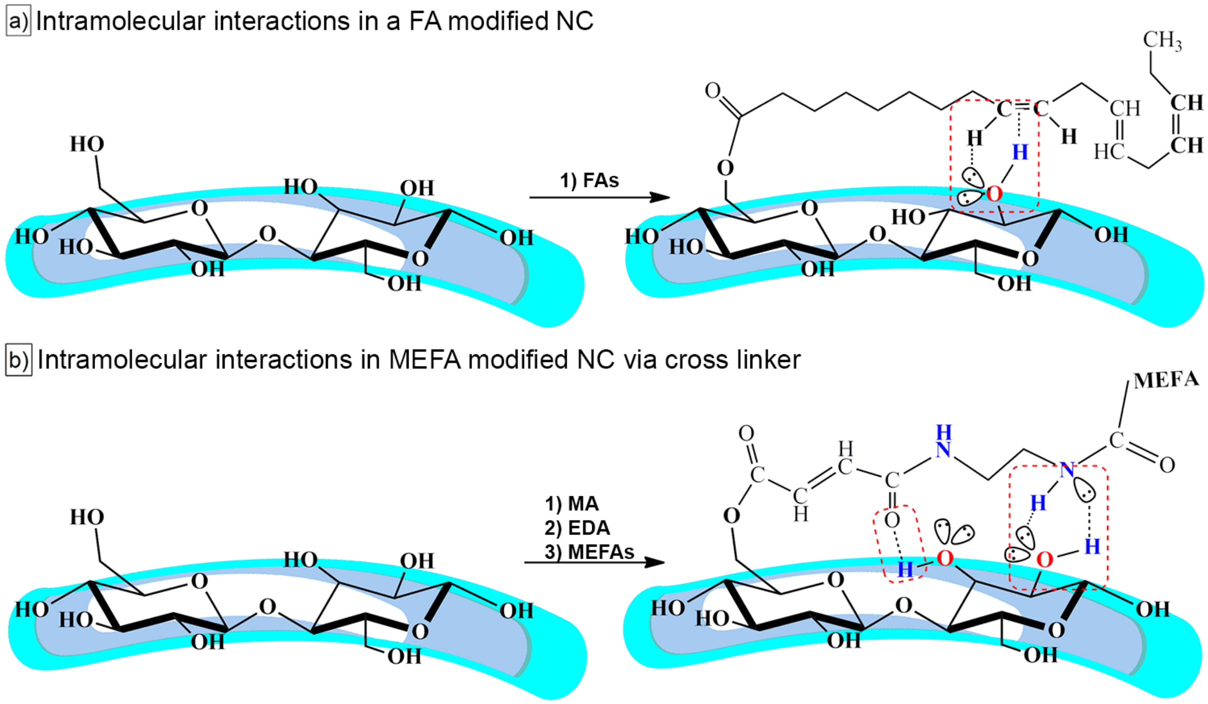

Fig. S2. Schematic overview of NC modification with a) FAs and b) MEFAs *via* cross-linker

2.6 Degree of substitution per anhydroglucose unit

The degree of substitution value (DS_{Gr}) per anhydroglucose unit (AGU) was determined by gravimetric measurements and calculated using the following equation (Almasi, Ghanbarzadeh, Dehghannia, Pirsa, & Zandi, 2015):

$$DS_{Gr} = (GAIN \times MW_{AGU})/(100 \times (MW_{ACID} - MW_{OH}) \quad \text{(S1)}$$

where GAIN (%) is the weight gain of NC after the esterification reaction calculated using the following equation:

$$GAIN = [100 \times m_3/(m_1 - m_2)\,] - 100 \quad \text{(S2)}$$

where m_1 and m_3 are the weights (g) of NC before and after the esterification with OA and FA isolated from SO and LO, respectively, and m_2 is the weight loss of NC after the reaction only with pyridine (all weights are calculated for an equivalent of 0.100 g of NC); MW_{AGU}, the molecular weight of one anhydroglucose unit of cellulose (162 g/mol); MW_{ACID}, the molecular weight of FAs with contribution of all typical FAs isolated from LO and SO (MW_{OA}=282.74 g/mol; MW_{LO}=278.86 g/mol; MW_{SO}=280.09 g/mol); and MW_{OH}, the molecular weight of one hydroxyl group (17 g/mol).

2.7 Supercritical CO_2 drying of NC

One of the major NC isolation challenges is to obtain dry nanosized NC of high thermal stability. During high temperature oven drying process with circulating air, water removal forces and temperature cause agglomeration of NC. The SCD of NC was carried out in a semi-batch Autoclave Engineers Screening presented on **Fig. S1**. This unit was designed for a small batch research runs with a maximum allowable working pressure of 41.3 MPa at 238 °C. Liquid CO_2 is supplied from CO_2 cylinder (T) with a siphon tube and pressurized with a high pressure liquid pump (LP).

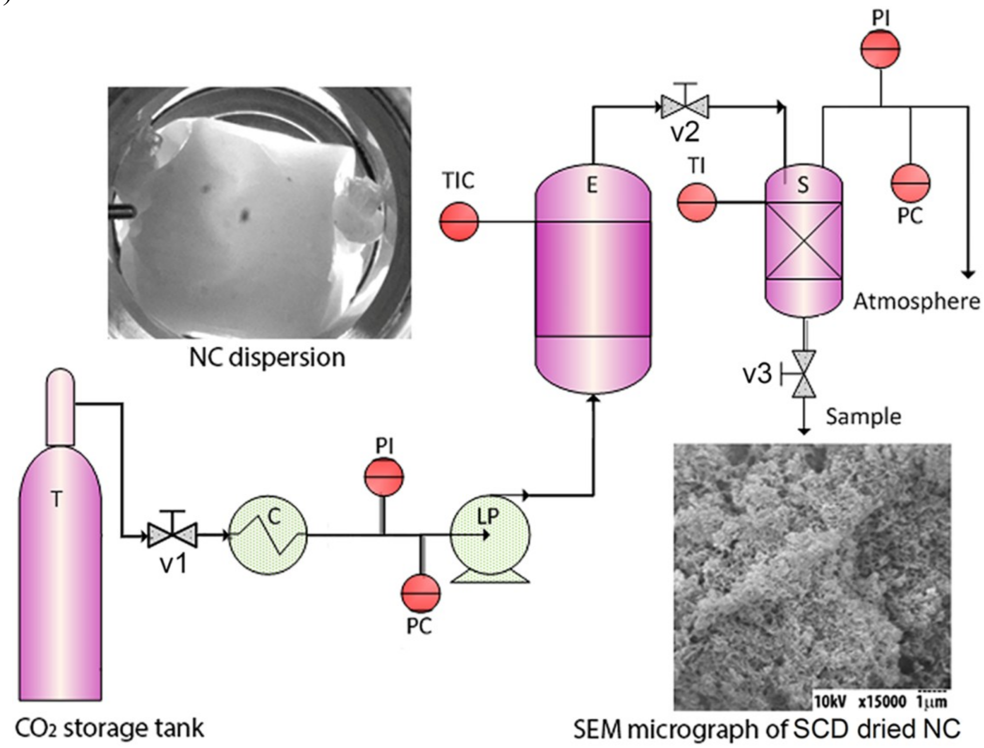

Fig. S3. Schematic presentation of the autoclave engineers screening system—T: CO_2 storage tank; C: cryostat; LP: high pressure liquid pump; E: extractor vessel; S: separator vessel (Glišić et al. 2010).

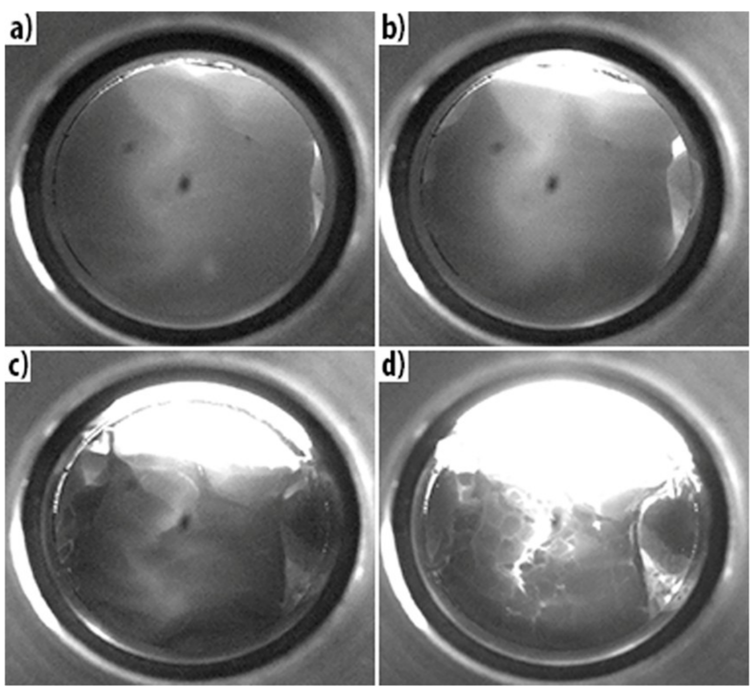

Fig. S4. Influence of $scCO_2$ on NC at 20 MPa and 40 °C; a) at the beginning static treatment, b) after static treatment, c) after 1 h, and d) after 2 h of dynamic SCD

2.8 Synthesis of UPe resin

UPe resins were synthesized from maleic anhydride and products of, obtained by poly(ethylene terephthalate) (PET) depolymerization with propylene glycol (PG) in the presence of tetrabutyl titanate catalyst (TBT). Following the glycolysis procedure described earlier in the literature (Rusmirović et al.,2015; Rusmirović et al.,2016), molar ratio of PET and PG used for glycolysis was 1:2.2 and reaction was maintained at 210°C. After completion of the glycolysis reaction, keeping inert atmosphere, mixture was cooled down to 90 °C and the Dean-Stark separator was assembled. MA (123 g, 1.25 mol) and 0.03 g of hydroquinone (HQ) dissolved in 2 ml of ethanol were charged, whereupon the mixture was heated to 115°C at constant temperature for 1 h. Afterwards, continuous temperature increase was achieved at a heating rate of 15 °C/h until 150 °C, when the toluene azeotropic removal of water was started. The temperature increase was continued until 210°C. The reaction was conducted until the acid number value decreased below 30 mg KOH/g, after which the resin obtained was cooled down to 120 °C and a solution of the 0.03 g HQ in 2 ml of methanol was added. After purification product by vacuum distillation the resin was dissolved in styrene (40 wt%).

2.10 Characterization methods

The structural analysis of the isolated NC was performed by FTIR (Bomem MB-102) spectroscopy, within a range of 400-4000 cm^{-1}, at a resolution of 4 cm^{-1} and in ten scan mode. The prepared procedure was consisted of sample homogenization with KBr (1 mas.% of sample) and compression of homogenized mass into pill using laboratory hydraulic press.

The hydroxyl value (*HV*) was determined using a conventional acetic anhydride/pyridine method (ISO 4326:1992) (International Organization for Standardization, 2012). The acid value (*AV*) was determined using a standard method ASTM D3644 (American Society for Testing and Materials, 2015). Ester value (*EV*) was determined using European quality standard for fatty acid methyl esters EN 14103 (European Committee for Standardization, 2003). Iodine value (*IV*) was determined by the Wijs method (Mscutcheon, 1940).

Microstructural (morphological) characterization was performed on a scanning electron microscope (SEM) JEOL/EO INSTRUMENT JSM-6390 and transmission electron microscope (TEM) JEM–1400.

Uniaxial tensile measurements of standard cured samples (standard dimension 60x10x4mm with narrowed neck area – 15x4x4mm) (ASTM D882) (American Society for Testing and Materials, 2009) were performed using an AG – X plus Universal testing machine, Shimadzu. All tests were performed at room temperature adjusted at crosshead speed of 0.5 mm/min.

On-line thermogravimetric–mass spectrometry analyses (TG–MS) was used to study the gases evolved during temperature-programmed heating of investigated samples. The experiments were performed using the TG/DSC 111 from Setaram, consisting of a quartz microreactor heated in a vertical furnace. An on-line mass spectrometer (MS, Thermostar from Pfeifer) was applied as the detector, the capillary-coupling system was used. Approximately 2 mg of each sample was heated from 25 up to 800 °C with a heating rate of 10 °C min^{-1}. The experiments were carried out in helium (flow rate 30 cm^3/min). Along with this temperature increase, the mass spectrometer was programmed to collect specific *m*/*z* values.

Particle sizes of all samples were measured in aqueous suspensions (0.1 wt.% suspension of unmodified VC and SCD dried NC and 0.01 wt.% suspension of FAs and MEFA m-NC) by dynamic light scattering using Horiba SZ-100 nano Partica analyzer.

The charge of the surface of particles was characterized through the zeta potential of aqueous suspensions (the same wt.% as well as suspensions used for determining particle size) which was measured using the same machine: the samples were injected into a disposable cell and a measurement of the particle electrophoretic mobility results in the calculated zeta potential. The zeta potential was also measured as a function of pH (from 5.5 to 1.0) which was adjusted by adding appropriate HCl solutions.

Raman spectra, recorded in the range 3400–100 cm^{-1}, were collected with a XploRA Raman spectrometer from Horiba Jobin Yvon. The system employed laser at 532 nm (maximum output power 20-25mW). All the measurements were realized using the spectrometer equipped with a 2400 g/mm grating. The Raman spectrometer was connected with an optical microscope equipped with a motorized stage.

NanoScope III A (Veeco, USA) microscope was used to study the morphology of unmodified NC suspension on the mica surface by atomic force microscopy (AFM), which operated in contact mode under ambient conditions. Silicon nitride probes were used. Samples were prepared by applying a 10µl of suspension on freshly clean mica plate.

3.1 Yields of NC obtained by acid hydrolysis

The yield of NC (%) obtained by acid hydrolysis of cotton with 64% H_2SO_4 was calculated according to equation S3 (Tang, Yang, Zhang, & Zhang, 2014). The achieved yield was 32% and applied procedure gave very stable colloidal water suspension which was attributed to negatively charged sulfate groups at the NC surface. Obtained results are in accordance with literature one (Ioelovich, 2012; Tang et al., 2014). It was also expected that, in first instance, hydrolytic reaction dominates, and in second both hydrolytic and sulfonation reactions contribute to production of sulfonated nanoparticle of smaller size and thus higher dispersion stability.

$$Yield\ (\%) = [((m_1 - m_2) \times V_1)/(m_3 \times V_2)] \times 100 \quad \text{(S3)}$$

Where m_1 is the sum of the total mass of vacuum dried NC and Erlenmeyer flask in which NC was dried (g); m_2 is the mass of the Erlenmeyer flask (g); m_3 is the mass of cotton used for NC isolation (g); V_1 is the total volume of NC suspension after dialysis (mL); V_2 is the volume of vacuum dried NC (mL). NC was vacuum dried at 50 °C to a constant weight.

3.5 Raman spectroscopy

$$X_{Raman} = [(I_{377}/I_{1091}) - 0.0286]/0.0065 \quad \text{(S4)}$$

3.7 Morphology analysis

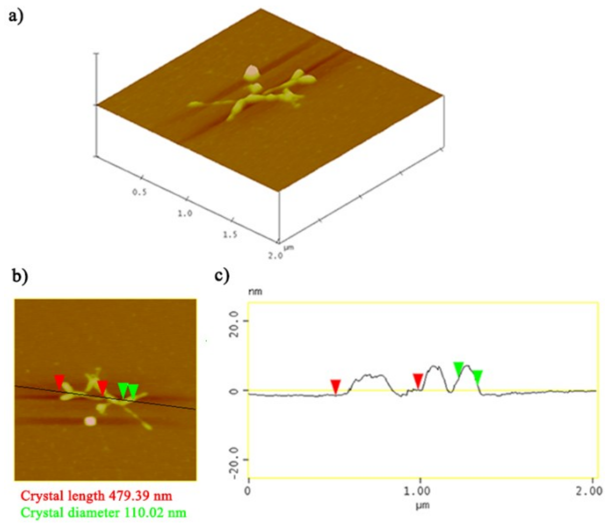

Fig S5. a) 3D AFM image, b) 2D S AFM image and height profiles (2 μm x 2 μm x 0.05 μm)

3.9 Electrokinetic phenomena

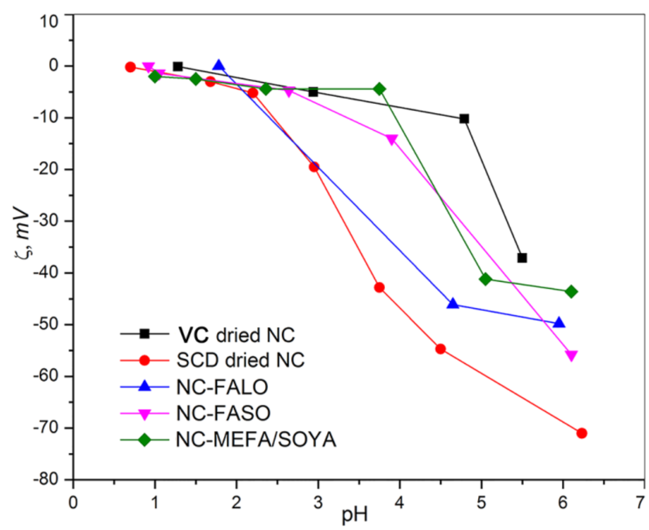

Fig. S6. The pH dependences of the ζ potential of unmodified and m-NC particles

3.10 Reinforcement effect of m-NC in UPe based nanocomposites

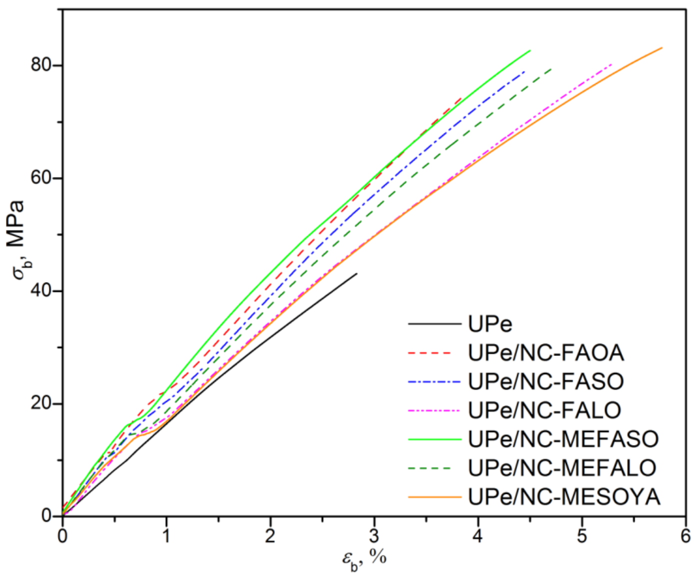

Fig. S7. Stress-strain curves of UPe/m-NC based composites

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